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Stem cells in sepsis and acute lung injury

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Abstract

Objective—Sepsis and acute lung injury continue to be major causes of morbidity and mortality worldwide despite advances in our understanding of pathophysiology and the discovery of new management strategies. Recent investigations show that stem cells may be beneficial as prognostic biomarkers and novel therapeutic strategies in these syndromes. This article reviews the potential use of endogenous adult tissue-derived stem cells in sepsis and acute lung injury as prognostic markers and also as exogenous cell-based therapy.

Data Sources—A directed systematic search of the medical literature using PubMed and OVID, with particular emphasis on the time period after 2002, was done to evaluate topics related to 1) the epidemiology and pathophysiology of sepsis and acute lung injury; and 2) the definition, characterization, and potential use of stem cells in these diseases.

Data Synthesis and Findings—When available, preferential consideration was given to prospective nonrandomized clinical and preclinical studies.

Conclusions—Stem cells have shown significant promise in the field of critical care both for 1) prognostic value and 2) treatment strategies. Although several recent studies have identified the potential benefit of stem cells in sepsis and acute lung injury, further investigations are needed to more completely understand stem cells and their potential prognostic and therapeutic value.

Keywords

stem cells; sepsis; ARDS; biomarkers; cell-based therapy

Sepsis, the tenth leading cause of death in the United States, is a significant public health problem. Acute lung injury (ALI) and acute respiratory distress syndrome (ARDS), severe forms of hypoxemic respiratory failure and frequent complications of sepsis (1), often result in prolonged mechanical ventilatory support with a mortality of approximately 30–50% (1). Despite years of research and recent advances in therapeutic strategies for these two diseases (2–4), morbidity, mortality, and healthcare expenditures remain high (1, 5).

Considerable research has identified several of the pathophysiological responses that occur when a host responds to a systemic infection. Proinflammatory cytokines such as tumor necrosis factor- α 3 and interleukin-6 (IL-6) characterize sepsis (6, 7) and ALI/ARDS (8–10), whereas coexisting anti-inflammatory pathways modulate the inflammatory response (11). An improved understanding of this pathophysiology has resulted in aggressive attempts to identify pathogenetically important biomarkers and new therapies. Both embryonic and adult tissue-derived stem cells have shown remarkable potential to repair and

regenerate various organs, including the lungs (12–14). For the purposes of this review, we will only discuss adult tissue-derived stem cells. Additionally, stem cells are able to mitigate injury and inflammation through paracrine mechanisms (12, 15, 16), and detecting their presence may help prognosticate survival (17, 18). These findings suggest that stem cells may offer novel approaches for both prognosis and cell-based therapies in sepsis and ALI/ARDS.

Stem Cell Classification

One of the most important characteristics of stem cells is their capacity for self-renewal and potential to change into cells of multiple lineages (19). Individual stem cells self-generate, undergoing continuous cell formation leading to a succession of cells that have progressively reduced capacity for self-generation until, ultimately, a lineage committed cell is formed. Until recently, the beneficial effects of stem cells were mostly attributed to their ability to incorporate into tissue (engraftment), differentiate into the appropriate cell type, and repair injured areas. Although engraftment may still occur with some stem cells, recent investigations propose that other mechanisms may be involved. For example, stem cells can exert paracrine effects with the capacity to generate chemokines and growth factors necessary for tissue repair.

Stem cells are broadly classified into adult tissue-derived vs. embryonic stem cells. Embryonic stem cells, derived from the inner cell mass of a developing blastocyst, are designated as pluripotent and can proliferate indefinitely without differentiation, potentially leading to the formation of neoplasms (20–22). In contrast, adult tissue-derived stem cells are also able to differentiate into a variety of adult tissues, but the fate of these cells seems to be somewhat restricted. For the purposes of this review, we discuss adult tissue-derived stem cells, focusing on mesenchymal stromal cells (MSC) and endothelial progenitor cells (EPC).

MSC can be isolated from multiple tissues (23, 24), but the best characterized source is the bone marrow. The small numbers present in the bone marrow usually necessitate expansion of these cells *in vitro*. Consensus minimal criteria to define human MSC include 1) selection for a plastic-adherent cell population in standard culture conditions; 2) expression of CD105, CD73, and CD90 and lack expression of CD45, CD34, CD14, CD11b, CD79α, CD19, or HLA-DR surface molecules; and 3) ability to differentiate into adipocytes, osteocytes, and chondrocytes *in vitro* (25, 26). In the lung, bone marrow-derived MSCs are able to engraft as type 1 and type 2 alveolar epithelial cells, endothelial cells, fibroblasts, and bronchial epithelial cells (12, 16). Why are MSC attractive for cell-based therapy? MSCs have immunomodulatory characteristics that augment tissue repair. In animal models of lung injury, administration of these cells attenuates the severity of the inflammatory response despite low levels of cell engraftment (12, 14). In addition, MSCs are not immunogenic; they have an innate ability to avoid detection by a recipient's immune system (27). MSCs inhibit lymphocyte proliferation (28, 29) expressing only intermediate levels of Major Histocompatibility Class I but not Major Histocompatibility Class II (30). Thus, Major Histocompatibility Classes are suitable for transplantation even between MSC-incompatible individuals.

In contrast to MSC, EPCs are a subtype of hematopoietic stem cells with the ability to only differentiate into mature endothelial cells. First discovered in 1997 (31), EPCs have been isolated from circulating mononuclear cells (31), bone marrow (32), and cord blood (33). EPCs migrate from bone marrow to peripheral blood where they contribute to vascular repair and new blood vessel formation. In contrast to MSCs, the phenotypic characterization of EPC is still uncertain, because there is no unique marker in animals or humans to define an EPC. Several culture methods have been described (31, 34–36), but typically the initial

population in cell culture is heterogeneous, making it difficult to determine the precursor cell that gives rise to EPC. Fluorescence-activated cell sorting can potentially identify more homogeneous EPCs using specific cell surface antigens (37), although this method is not ideal because there are a number of overlapping markers among EPCs and other hematopoietic cells (31, 38, 39). EPCs have been extensively studied as biomarkers of cardiovascular disease and as cell-based therapy for repair of damaged vasculature (35, 40). However, similar to MSC, EPC investigators continue to speculate an alternate mechanism besides engraftment, namely paracrine effects given the lack of evidence of long-term engraftment into newly formed vessels (41, 42).

Stem Cell Mobilization

The severity of the inflammatory response in sepsis may cause organ dysfunction, a primary determinant of survival (43), but the development of organ dysfunction is variable and not predicted by clinical or physiological variables (44–47). Low doses of endotoxin activate macrophages (48) and release a variety of chemoattractant factors and signaling cascades. These systemic signals mobilize stem cells from the bone marrow and recruit them to injured sites at which point they may differentiate into a variety of tissue-specific cell types (12, 14, 25, 49, 50) and modify the immune response (27, 51). Pulmonary epithelial and endothelial cells of donor origin have been identified in the injured lung after hematopoietic stem cell and lung transplant in both murine and human studies (52–54), suggesting that stem cells have the ability to home to injured organs (14, 52, 55), modulate the immune response, and repair damaged tissue (29, 56–58). Mobilization of stem cells in both preclinical and clinical models of sepsis and ALI also correlates with clinical outcomes (17, 18, 59).

There is enthusiasm to study stem cells in human diseases. An assortment of clinical trials have used stem cells in cardiovascular diseases (35, 40, 57, 60), heart failure (61), pulmonary hypertension (62), graft-versus-host disease (13), and cerebrovascular disease (63). However, until recently, stem cells were not extensively studied in sepsis and ALI/ARDS.

Stem Cells in Sepsis

In recent years, several studies have been published with both preclinical and clinical data regarding the prognostic and therapeutic benefits of stem cells in sepsis.

Prognostic Implications of Stem Cells in Sepsis

Stem cells, in particular EPC, have been investigated as prognostic biomarkers in sepsis (Table 1). In 2001, Mutunga et al identified by indirect immunofluorescence a significant increase in circulating endothelial cells in a small group of patients with sepsis and septic shock compared with intensive care unit control subjects or healthy people (64). Increasing numbers of endothelial cells were found even in patients with sepsis without shock, suggesting that endothelial damage preceded the development of organ failure (64). In addition, the number of circulating endothelial cells was higher in patients who died of septic shock compared with survivors, supporting the view that the magnitude of increase in circulating cell numbers in sepsis maybe related to the severity of vascular injury (64). Although this study detected circulating endothelial cells, as opposed to EPCs, it provided direct evidence for endothelial dysfunction in humans during septic shock. Subsequently, Tsaganos et al (65) studied 44 patients with ventilator-associated pneumonia and sepsis, a more homogeneous population than previous studies, and observed that survival time was decreased for patients with $310/\mu\text{L}$ of CD34/CD45 cells on day 1 compared with those patients with $<310/\mu\text{L}$ of cells. These results were similar to the findings of Mutunga et al

(64) because mortality was associated with greater number of circulating CD34/CD45 progenitor cells.

Rafat et al (18) measured levels of circulating EPC identified by fluorescence-activated cell sorting using antibodies against CD34, CD133, and VEGFR-2 in a cohort of 32 patients within 48 hrs after sepsis onset. Like in the previous studies, the number of circulating EPCs was significantly higher in patients with sepsis vs. nonseptic intensive care unit patients and healthy control subjects (18). The number even remained elevated 5 days after the initial sampling. In addition, high numbers of circulating EPCs correlated with elevated serum levels of vascular endothelial growth factor, granulocyte monocyte-colony-stimulating factor, and erythropoietin (18). However, in contrast to previous studies, Rafat et al (18) found that sepsis survivors had significantly greater numbers of EPCs than nonsurvivors. Similarly, Burnham et al recently studied EPC in a cohort of patients with sepsis and ALI. EPCs were assessed by a colony-forming unit (CFU) cell culture assay in which peripheral blood mononuclear cells were isolated using Ficoll density-gradient centrifugation and cultured in EPC media. Analysis revealed that those with CFU count $\geq 48/\mu\text{L}$ had an overall better survival compared with those with CFU count $<48/\mu\text{L}$ (66). Our group at Emory University recently studied EPC, also quantified by a similar CFU culture assay, in a cohort of patients with sepsis as a biomarker for organ dysfunction, a major determinant of mortality in sepsis (Cribbs S, Sutcliffe D, Taylor WR. et al: Circulating endothelial progenitor cells inversely associate with organ dysfunction in sepsis; unpublished data). In this study, EPC CFU counts were significantly lower in patients with sepsis compared with critically ill and healthy control subjects; however, similar to previous studies (18, 66), higher EPC CFU counts correlated with improvement in organ dysfunction (as measured by Sepsis-related Organ Failure Assessment score) regardless of shock or survival status.

Disparate results in these studies may be explained by differences in the techniques used for EPC identification. The flow cytometry technique used by Rafat et al contrasts to the CFU cell culture technique used by Burnham et al and our group. Cell-culturing techniques may offer a measurement of EPC functionality that fluorescence-activated cell sorting analysis does not; however, these techniques may identify different progenitor cell types. Given the lack of consensus on definitive identification of EPC, which technique to use and which cells to label as EPC remain ambiguous. In addition, variability in these sepsis populations could also explain observed differences. Bone marrow suppression and vascular dysfunction will vary in severity from patient to patient, which can ultimately result in variable EPC measurements.

Therapeutic Potential of Stem Cells in Sepsis

Several studies evaluating cell therapy in sepsis have been done regarding progenitor cells other than EPC, namely bone marrow-derived MSC (Table 2). The group of Xu et al (16) hypothesized that bone marrow-derived MSC, expanded *in vitro* and delivered to mice, would inhibit the acute inflammatory response from endotoxin, effectively preventing ALI. Lungs from mice receiving endotoxin but not MSC showed vascular congestion and increased cellularity, attributable mostly to neutrophils, whereas those receiving intravenous MSC plus endotoxin resembled control mice (16). In addition, MSC moderated the increase in systemic proinflammatory cytokines but did not alter serum concentrations of the anti-inflammatory cytokine, IL-10 (16).

More recently, MSCs have been evaluated in other murine models of sepsis. Mice given bone marrow-derived MSCs at the time of surgery for cecal ligation and puncture lived longer than untreated animals, particularly when administered between 24 hrs before to 1 hr after cecal ligation and puncture, and kidney and liver function were markedly improved (67). Similar to others, this group found that tumor necrosis factor- α and IL-6

concentrations were reduced in treated mice, whereas IL-10 levels increased 3 hrs after cell infusion. In addition, treatment with MSC reduced peritoneal, renal, and liver vascular permeability that occurs in this model of peritoneal sepsis. The authors also discovered that although lymphocyte populations of T, B, and natural killer cells did not mediate the effect of MSC on cecal ligation and puncture, MSCs were no longer effective in mice lacking monocytes, macrophages, or IL-10, suggesting that injected MSCs may interact with circulating immune cells to reprogram the host immune response (67). Similarly, Gonzalez-Rey et al (68) discovered that treatment with human or murine adipose-derived MSCs in a sepsis model increased survival in mice. A recently published study treating peritoneal sepsis in mice also demonstrated that intravenous bone marrow-derived MSCs were effective in reducing mortality, even when compared with control mice given antibiotics (69). In these studies, IL-10 was not an important mediator of benefit, although the MSCs did have a marked effect on reducing proinflammatory cytokines like in prior studies.

Stem Cells in ALI

Recent advancements with bone marrow-derived MSC and EPC have also been made in preclinical studies of ALI/ARDS. Some investigators have demonstrated the ability of stem cells to repair damaged lung epithelium and endothelium (14, 52, 55, 59) and have correlated these cells to clinical outcomes.

Prognostic Implications of Stem Cells in ALI

Burnham et al hypothesized that increased levels of circulating EPCs in the first 72 hrs of ALI would be associated with an improved outcome. The authors found that patients with ALI/ARDS had significantly greater EPC CFU counts compared with healthy control subjects and that patients with ALI with an EPC CFU count ≥ 35 per volume had a greater 28-day survival compared with those patients with an EPC count <35 per volume (17). They concluded that adequate mobilization of EPC from the bone marrow in ALI/ARDS could contribute to repair and recovery of damaged pulmonary endothelium (17), conferring a survival benefit. Furthermore, EPC counts were similarly elevated in the 44% of subjects with septic shock (17), again suggesting that vascular damage induces release of EPC into the circulation (Table 1).

Therapeutic Potential of Stem Cells in ALI

The therapeutic effects of EPCs have also been demonstrated and regeneration of pulmonary endothelium may be a potential therapeutic target for patients with ALI/ARDS (Table 2). Kahler et al reported that EPCs are integrated into the endothelial tissue of a transplanted lung with ALI, although virtually no cells were found in the healthy lung (52), illustrating that EPC home to areas of vascular injury. Lam et al (70) studied the effects of autologous EPC transplantation on pulmonary endothelial regeneration in a rabbit model of ALI and showed that endothelial dysfunction in the pulmonary artery was significantly attenuated in rabbits treated with EPCs. The cells potentiated the relaxation response to acetylcholine in pulmonary arteries, reducing lung water content and hyaline membrane formation. The therapeutic benefits of EPC in this model were most likely derived from an effect on re-endothelialization of the damaged pulmonary artery wall and alveolar–capillary membrane. Most recently, Mao et al (71) isolated EPCs from male donor bone marrow and expanded them *in vitro* before infusing them into female rats with lipopolysaccharide-induced ALI. Using Y-chromosome *in situ* hybridization and reverse transcription–polymerase chain reaction to confirm engraftment, they found that rats treated with EPCs had reduced pulmonary edema, hemorrhage, and hyaline membrane formation in addition to an increased survival rate (44% vs. 81%, $p = .03$) compared with saline-treated controls (71). EPCs were not detected in lungs without lipopolysaccharide, again implying that injured lung cells

produce chemoattractant factors that allow EPC to mobilize to the injury site. Furthermore, anti-inflammatory cytokines were increased in EPC-treated rats (71). Kahler et al and Mao et al obtained EPCs from bone marrow, whereas Lam et al used peripheral blood. However, in all three studies, mononuclear cells were isolated by density-gradient centrifugation, cultured in endothelial growth medium, and EPC were harvested in 7–10 days. Additionally, EPCs were characterized by staining with acetylated low-density lipoprotein and isolectin. These findings indicate that the benefits of EPC can be explained by both repair of damaged endothelium and reduction in inflammation.

The immunologic tolerance of MSCs, along with their anti-inflammatory characteristics, makes them potentially attractive for cell-based therapy of ALI/ARDS as well. Several animal studies have shown that exogenously administered MSCs augment tissue repair; these effects appear to be mediated by soluble factors produced by MSC, because levels of engraftment are too low to account for their protective effects (12, 16). Administration of MSCs attenuated the acute inflammatory response to lipopolysaccharide and protected the lung from injury and pulmonary edema in several preclinical models of ALI (12, 14, 16, 72–75), even conferring a survival benefit (12). Infusion of MSCs completely attenuated neutrophil infiltration in the lung between 6 hrs and 48 hrs (16). In this study, further investigation showed that MSCs migrated in response to lipopolysaccharide when placed in a co-culture environment, again suggesting that chemoattractant factors from the injured lung initiated chemotaxis of MSC (16). Gupta et al found that intrapulmonary delivery of MSC improved survival from endotoxin-induced ALI in mice compared with saline-treated control mice (80% vs. 42%; $p < .01$). Fibroblasts and apoptotic MSCs were also used as controls and showed no effect (12). Mice given MSCs had less pulmonary edema at 48 hrs compared with mice that received the fibroblast cell line or apoptotic MSC, and there was significant histologic improvement in the severity of lung injury after MSC administration despite <5% engraftment at 48 hrs after injury. The beneficial effects with MSCs appear to be mediated by a shift from a proinflammatory to an anti-inflammatory response to endotoxin (12), an effect also noted by Xu et al (16).

In addition to murine models of ALI, this group also tested the ability of human allogeneic MSCs to resolve pulmonary edema in an *ex vivo* perfused human lung preparation injured by endotoxin (76). After *Escherichia coli* endotoxin-induced ALI, treatment with either allogeneic human MSC or human MSC-conditioned medium reduced pulmonary edema, improved lung endothelial barrier integrity, and normalized alveolar epithelial fluid transport (76) with concomitant reductions in inflammatory cell infiltration and lung septal thickening. Previous animal studies have shown that alterations in pro- and anti-inflammatory cytokines may account for some of these beneficial effects (12), but anti-inflammatory cytokines were not elevated in this *ex vivo* human model. However, the levels of monocytes and neutrophils were lower compared with their previous animal model (12), leading the authors to believe that these low levels may represent an early anti-inflammatory effect or lung recovery (76). In addition, the authors also tested the ability of MSC to secrete keratinocyte growth factor, which has been shown to reduce ALI in small animal models of pulmonary edema (77, 78). They found that MSCs pretreated with keratinocyte growth factor small interfering RNA resulted in an 80% reduction in therapeutic effect. This effect was restored by the addition of recombinant keratinocyte growth factor, suggesting that secreted keratinocyte growth factor is an important paracrine soluble factor that mediates some of the beneficial effect of MSC on alveolar fluid clearance (76).

The combination of gene plus cell therapy has also been studied in ALI. Limitations of stem cells such as recruitment, engraftment, and possible lung sequestration make gene transduction an attractive alternative approach because this dual strategy allows for direct targeting of the lung- and site-specific release of cellular products. Mei et al (73) evaluated

the effect of MSCs in combination with a plasmid containing human angiopoietin 1 gene on lung injury in a murine model of ALI. Although MSC infusion to the pulmonary circulation partially prevented lung inflammation, as reflected by reductions in neutrophil counts and proinflammatory cytokines in bronchoalveolar lavage, treatment with plasmid containing human angiopoietin 1 gene-transfected MSCs resulted in further improvement in alveolar inflammation and permeability (73). This same group reported that EPCs engineered to overexpress endothelial nitric oxide synthase were more effective than EPC alone in a monocrotaline model of vascular injury, producing significant reduction in pulmonary hypertension (79). Similarly, Xu et al (80) found that MSC transduced with the angiopoietin-1 gene exerted a beneficial therapeutic effect in treatment of lipopolysaccharide-induced lung injury, whereas administration of MSC or angiopoietin-1 alone had little effect. These preclinical studies suggest that combining stem cell-based therapy with gene therapy might have additive therapeutic effects in patients with ALI.

CONCLUSIONS

Considerable progress has been made in the last decade regarding management strategies in sepsis and ALI/ARDS. Despite advances in supportive care, more work needs to be done to improve survival and quality of life and to reduce morbidity and healthcare costs of critically ill patients. Stem cells have shown significant promise, both as biomarkers to prognosticate organ dysfunction and mortality and as potential novel therapies to treat organ failure, including ALI. However, there are many complexities with using stem cells for prognostication or for cell-based therapy. In particular, there are significant challenges in accurate identification of cells, the precise collection of cells, mobilization of adequate numbers of cells, and optimal method of delivery. The future of cell-based therapy may focus on the stimulation of other cells (growth factors, cytokines, and various other hematopoietic elements) that not only recruit and facilitate endothelium and epithelium formation, but may have anti-inflammatory properties as well (81–84). Further research is necessary to identify these mechanisms so that novel therapeutic modalities can be studied in critically ill patients.

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Table 1

Stem cells as prognostic biomarkers in critically ill patients

Author	Date	Cell Type	Disease	Patients (No.)	Clinical Outcome	p
Mutunga et al	2001	Endothelial cells	Sepsis	Septic shock (15) Sepsis (8) ICU control (9) Healthy (11)	Positive correlation with mortality	.026
Burnham et al	2005	Endothelial progenitor cells (colony-forming unit)	ALI/ARDS	ALI (45) ICU control (10) Healthy (7)	Positive correlation with survival	<.04
Tsaganos et al	2006	CD34/CD45 cells	Ventilator-associated pneumonia and sepsis	Sepsis (44) Healthy (8)	Positive correlation with mortality	.022
Rafat et al	2007	Endothelial progenitor cells (FACS)	Sepsis	Sepsis (32) ICU control (15) Healthy (15)	Positive correlation with survival	<0.0001
Burnham et al	2009	Endothelial progenitor cells (colony-forming unit)	Sepsis and ALI/ARDS	ALI (65) Severe sepsis (17) Healthy (5)	Positive correlation with survival	.06
Cribbs et al	Unpublished data	Endothelial progenitor cells (colony-forming unit)	Sepsis	Sepsis (86) ICU control (37)	Positive correlation with <i>improved</i> organ dysfunction	<.05

ALI, acute lung injury; ARDS, acute respiratory distress syndrome; ICU, intensive care unit; FACS, fluorescence-activated cell sorting.

Table 2

Preclinical therapeutic studies of stem cells in sepsis and ALI/ARDS

Author	Date	Cell Type	Disease	Clinical Outcome With Stem Cells
Rojas et al	2005	Bone marrow-derived MSC	Bleomycin-induced ALI	Histologic decrease in lung injury Increase in GM-CSF and G-CSF Decrease in proinflammatory cytokines
Gupta et al	2007	Bone marrow-derived MSC	Endotoxin induced ALI	Histologic decrease in lung injury Increase survival Decrease in excess lung water and BAL protein Decrease proinflammatory cytokines Increase anti-inflammatory cytokine
Xu et al	2007	Bone marrow-derived MSC	Endotoxin induced ALI	Histologic decrease in lung injury Decrease in excess lung water Decrease proinflammatory cytokines Increase anti-inflammatory cytokine
Kahler et al	2007	Bone marrow-derived EPC (cultured <i>in vitro</i>)	ALI	Integration of EPC into tissue of injured lung
Ortiz et al	2007	Bone marrow-derived MSC and MSC-conditioned medium	Bleomycin-induced ALI	Decrease proinflammatory cytokines Decrease BAL protein
Zhao et al	2008	Bone marrow-derived MSC	Bleomycin-induced ALI	Histologic decrease in lung injury Decrease in laminin and hyaluronan in BAL Decrease proinflammatory cytokines
Lam et al	2008	Peripheral blood EPC (cultured <i>in vitro</i>)	ALI	Histologic decrease in lung injury Decrease endothelial dysfunction in pulmonary artery Decrease excess lung water
Nemeth et al	2008	Bone marrow-derived MSC	Sepsis	Improve survival and organ dysfunction Decrease proinflammatory cytokines Increase anti-inflammatory cytokines
Gonzalez-Rey et al	2009	Adipose-derived MSC	Sepsis	Histologic and clinical decrease in colitis Improve survival and organ dysfunction Decrease proinflammatory cytokines Increase anti-inflammatory cytokines
Lee et al	2009	Human allogeneic MSC and human MSC-conditioned medium	ALI	Histologic decrease in lung injury Decrease excess lung water Restored alveolar fluid clearance
Mei et al	2010	Bone marrow-derived MSC	Sepsis	Histologic decrease in lung injury Decrease BAL protein Improve survival in setting of antibiotics Improved organ dysfunction Decrease proinflammatory cytokines
Gene plus cell therapy				
Mei et al	2007	Syngeneic MSC with and without <i>ANGPT1</i> gene	Endotoxin induced ALI	Histologic decrease in lung injury Decrease in inflammatory cells in BAL Near complete reversal of increased lung permeability with gene transfection
Xu et al	2007	Bone marrow derived MSC with and without <i>Ang1</i> gene	Endotoxin induced ALI	Further histologic decrease in lung injury with gene transfection vs. MSC alone Further decrease excess lung water and BAL protein vs. MSC alone Further decrease in proinflammatory cytokines

ALI, acute lung injury; ARDS, acute respiratory distress syndrome; FACS, fluorescence-activated cell sorting; MSC, mesenchymal stromal cell; EPC, endothelial progenitor cell; GM-CSF, granulocyte-monocyte colony-stimulating factor; BAL, bronchoalveolar lavage.