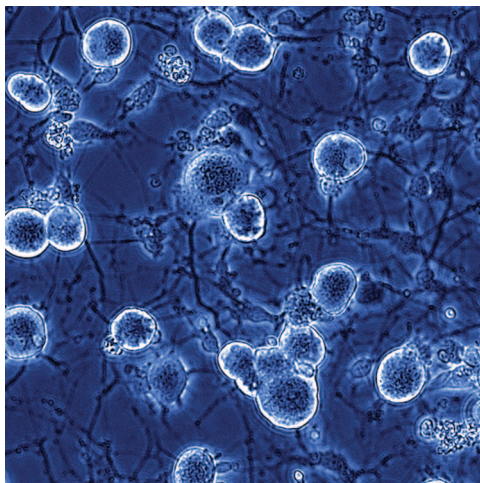


Cell-based Therapy for Acute Lung Injury

Are We There Yet?

ACUTE lung injury (ALI) and acute respiratory distress syndrome remain one of the most common causes of acute respiratory failure in critically ill patients.¹ Despite extensive research, current treatment options remain primarily supportive with lung protective ventilation and a fluid conservative strategy. Recently, the potential use of cell-based therapy has generated considerable interest for the treatment of lung diseases, including ALI.² In this month's issue of *ANESTHESIOLOGY*, Cao *et al.* add to the literature by studying the potential therapeutic role of circulating autologous endothelial progenitor cells (EPC) in endotoxin-induced ALI in rabbits.³

Much of the initial interest in cell-based therapy for treatment of ALI originated from the multipotent nature of bone-marrow-derived cells. In 2001, Krause *et al.*⁴ reported that a single bone-marrow-derived hematopoietic stem cell in mice could give rise to cells of multiple different organs, including the lung. The authors reported up to 20% engraftment of bone-marrow-derived cells in the lung, including epithelial cells, from a single hematopoietic precursor. This report stimulated additional investigations into the possibility that adult bone-marrow-derived stem cells might be able to regenerate the lung epithelium and/or endothelium. Initially, the regenerative potential of bone-marrow-derived EPC was studied predominantly in models of endothelial injury from cardiovascular disease, particularly after myocardial infarction or pulmonary arterial hypertension.⁵⁻⁹ More recently, several investigators demonstrated an improvement in pulmonary alveolar-capillary barrier function in lung injury models, including oleic acid in rabbits and endotoxin in rats after EPC therapy.^{10,11} However, subsequent



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the damaged pulmonary vascular wall by intercalating of the EPC into the injured capillaries or immunomodulation of the inflammatory and oxidant responses, including release of the antiinflammatory cytokine IL-10 and induction of higher concentrations of superoxide dismutase. The authors studied the trafficking of the EPC to the lung with immunohistochemistry and fluorescence-conjugated cell tracers for 48 h. However, because of the short time period of injury studied and the fact that the intravenous infusion of bone-marrow-derived cells typically are initially trapped in the pulmonary microcirculation, additional studies are needed to determine the contribution of engraftment in the therapeutic response of EPC. Cao *et al.* also identified the immunomodulatory properties of EPC as another potential therapeutic mechanism underlying the beneficial effect of EPC in lung injury.

The article by Cao *et al.* has several limitations. First, the authors were not able to identify the mechanisms of benefit

studies indicate that the level of engraftment of bone-marrow-derived cells in lung injury was low, with observed rates of less than 5%.¹²⁻¹⁶

In their study, Cao *et al.* found that intravenous infusion of EPC 4 h after the induction of endotoxin-induced injury significantly improved oxygenation (ratio of partial pressure of oxygen to fraction of inspired oxygen ratio) and histologic indices of lung injury, including the infiltration of polymorphonuclear cells, the extent of hyaline membrane formation and hemorrhage, and the lung wet-to-dry ratio as a measure of pulmonary edema at 48 h. EPC infusion suppressed the concentrations of the inflammatory cytokine, IL-1 β , and an adhesion molecule, ICAM-1, and also reduced concentrations of reactive oxygen species, nitric oxide and malondialdehyde. The authors speculated that the therapeutic effect of EPC could be attributable to endothelial repair of

Photomicrograph: Courtesy of Brian Wainger, M.D., Ph.D.

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◆ This Editorial View accompanies the following article: Cao J-P, He X-Y, Xu H-T, Zhou Z, Shi X-Y: Autologous transplantation of peripheral blood-derived circulating endothelial progenitor cells attenuates endotoxin-induced acute lung injury in rabbits by direct endothelial repair and indirect immunomodulation. *ANESTHESIOLOGY* 2012; 116:1278-87.

of EPC in these studies but provided circumstantial data suggesting engraftment or antioxidant or antiinflammatory effects. Second, an autologous source of the EPC was used for therapy, suggesting that an allogeneic source may cause an immune reaction in the host in a clinical setting. It is unclear if EPC are immunoprivileged in a manner similar to that of other adult stem cells, lacking in major histocompatibility complex I or II antigen expression, and able to evade a host response. Patients with ALI or acute respiratory distress syndrome usually experience rapid lung injury, so it would not be practical to harvest and culture circulating EPC from the patient before clinical use. In addition, the underlying causes of ALI, including infection, might alter the phenotype or number of circulating EPC or EPC from the bone marrow. For example, Burnham *et al.* found a higher number of colony-forming units of EPC from patients with ALI compared with healthy control subjects, and in patients with ALI, an increased number of circulating EPC were associated with improved survival, suggesting that circulating EPC were mobilized from the bone marrow to replenish the injured endothelium.¹⁷ Would these circulating EPC behave in a manner similar to that of circulating EPC or EPC from the bone marrow from otherwise healthy patients?

Finally, would EPC be the most effective cellular therapy for ALI. Another bone-marrow-derived adult stem or progenitor cell, mesenchymal stem cells (MSC), has been studied extensively in lung injury models. MSC secrete multiple paracrine factors that can regulate endothelial and epithelial permeability, decrease inflammation, enhance tissue repair, and inhibit bacterial growth.¹⁸ In preclinical animal models and in an isolated perfused human lung preparation, MSC have been effective in both endotoxin and live *Escherichia coli* bacteria pneumonia-induced lung injury, in part through the secretion of growth factors, such as keratinocyte growth factor, and antiinflammatory cytokines, IL-10.^{19–21} There are currently more than 200 clinical trials registered with *clinicaltrials.gov* testing MSC in a variety of disorders, including graft *versus* host disease, Crohn's disease, acute myocardial infarction, and acute kidney failure. More recently, MSC have been tested in a clinical trial for chronic obstructive pulmonary disease and bronchopulmonary dysplasia.

In addition, endogenous adult human lung stem cells recently were described with reparative and regenerative properties. Kajstura *et al.* reported a human c-KIT-positive adult lung stem cell that was clonogenic and able to regenerate the architecture of the lung bronchiole, alveoli, and arteriole after cryoablation injury in mice.²² However, the study needs to be replicated to determine the translational potential of these cells for acute or chronic lung diseases. Chapman *et al.* discovered a subpopulation of mouse alveolar epithelial cells expressing the laminin receptor $\alpha 6\beta 4$ that was capable of forming CC10-positive airway-like and SPC-positive sacular structures in a novel *in vivo* embryonic lung organoid assay.²³ Surprisingly, in a bleomycin mouse model of lung

injury, the authors found that the majority of alveolar type II cells formed after injury were not derived from preexisting type II cells but from these $\alpha 6\beta 4$ alveolar epithelial cells.

In conclusion, despite its limitations, the study by Cao *et al.* raises several important questions concerning the use of cell-based therapy for lung injury. What is the contribution of engraftment (if any) and of immunomodulation by the release of paracrine effector molecules or microvesicles²⁴ in the therapeutic effect of stem or progenitor cells? What is the most effective progenitor or stem cell to test clinically? In preclinical animal and human models of lung injury, both progenitor and adult stem cells have been shown to be effective in short-term models of ALI. In the clinical syndrome, ALI–acute respiratory distress syndrome, depending on the etiology and associated comorbidities, mortality rates range from 20 to 40%. Thus, innovative therapies are needed. In the future, well-designed clinical trials are warranted to test cell-based therapies for safety and efficacy.

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