Modified Hydrogels to Enhance Cellular Therapy for AKI: A Translational Challenge

Anna Gooch and Christof Westenfelder
Division of Nephrology and Hypertension, University of Utah and George E. Wahlen VA Medical Centers, Salt Lake City, Utah

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Numerous preclinical studies have demonstrated that the treatment of various models of AKI with either bone marrow-derived mesenchymal stromal or stem cells (MSCs) or their adipose-derived equivalent (ASCs) affords significant renoprotection and stimulates repair and functional recovery through the complex trophic and anti-inflammatory paracrine actions of these cells, and not through significant differentiation into tubular cells and engraftment.1–9 A phase 1 clinical trial (NCT00733876) demonstrated the safety and preliminary efficacy of allogeneic MSCs, administered into the suprarenal aorta of cardiac surgery patients at high risk for postoperative AKI.8,9 In this trial, cells were infused immediately after the study subjects were taken off cardiopulmonary bypass.8,9 However, in a subsequent phase 2 clinical trial (NCT01602328), MSCs were similarly administered but 1–2 days after a cardiac surgery-induced episode of AKI, diagnosed by a rise in serum creatinine. In this trial, outcomes (dialysis dependency, mortality, renal function) were worse in the treatment group than in the control group, and the trial was terminated because of “futility.”10 The numbers of MSCs in the stagnant microcirculation of the kidney, which occurs after most acute renal insults, further compromised renal function hemodynamically, and also reduced the intrarenal delivery of adequate numbers of renoprotective MSCs. Before this highly promising and effective cell-based therapy is abandoned, a scientifically based clinical trial, guided by the use of sensitive, early diagnostic biomarkers of AKI, such as Kim-1, IL-18, NGAL, SDF-1, or others, after a potential episode of AKI, is warranted.9,11

Mindful of the predominantly paracrine mode of actions of MSCs or ASCs in experimental AKI (antiapoptotic, anti-inflammatory, antithrombotic, antioxidant, mitogenic, angiogenic, and vasculoprotective), various modifications of this therapy have been used, primarily wishing to potentiate their organ-protective activity by boosting their homing to the injured kidney and by prolonging their intrarenal resident time.12 The intrarenal or remote release by MSCs of various growth factors such as IGF-1, hepatocyte growth factor (HGF), EGF, basic fibroblast growth factor, vascular endothelial growth factor, and others combined with these cells’ robust anti-inflammatory actions mediates the potent organ protective and repair-stimulating effects of MSCs.9 In the normal kidney, IGF-1 is expressed in collecting duct cells, and its receptors in the proximal tubule.13–15 Following AKI, IGF-1 is upregulated in the injured proximal tubule, and its administration to animals with AKI has been shown to increase the GFR and to improve recovery of renal function.13–16 The peripheral actions of IGF-1 are further modified by six IGF binding proteins, some of which are expressed both in the kidney and in MSCs.15,17 Preconditioning MSCs with IGF-1 enhances their therapeutic efficacy in experimental AKI by augmenting their homing to the injured kidney.18 Furthermore, ASCs and MSCs express IGF-1 and secrete exosomes that contain mRNA for the IGF-1 receptor that, when transferred to and translated in proximal tubular cells, contributes to their IGF-1-mediated renoprotective efficacy.16,19 Based on strong preclinical data that showed excellent renoprotective efficacy of IGF-1 in otherwise healthy animals with AKI, two major clinical trials were conducted in which study subjects with various comorbidities and AKI were treated with IGF-1, given up to 7 days after a renal insult.20,21 Not entirely unexpectedly, both trials showed no beneficial effects of this therapy, again illustrating that preclinical data that are obtained in healthy animals cannot readily be translated into the clinical situation where patients are at high risk for AKI because they have significant comorbidities. It obviously follows from this that novel therapies for AKI and other renal diseases must be shown to be effective in animals with clinically representative comorbidities before they are tested clinically, which has, however, only rarely been done. Importantly and uniquely, the pleiotropic anti-inflammatory and trophic actions of MSCs have the capacity to sequentially address all pathophysiologic components of AKI, which single factor interventions cannot.9

In the current issue of the Journal of the American Society of Nephrology, Feng et al. incorporate several of the above-discussed concepts and considerations related to the treatment of AKI with MSCs or ASCs.22 They specifically demonstrate that the combined injection of ASCs and IGF-1–modified hydrogel into the renal parenchyma improves their therapeutic efficacy in a single kidney model of ischemia-reperfusion injury AKI in mice. The utilized thermosensitive, biocompatible chitosan hydrogel was modified by covalently binding the 12-amino acid, bioactive C-domain of IGF-1 (CS-IGF-1) to the hydrogel in order to generate a stem cell niche-like environment that was postulated to enhance in vitro survival and proliferation of ASCs through paracrine actions. In vitro, the expression levels of IGF-1, HGF, and EGF by ASCs coated with modified hydrogel increased significantly, contributing to enhanced cell proliferation and antiapoptotic actions. The antiapoptotic activity of hydrogel embedded ASCs correlated well with...
a generalized downregulation of proapoptotic genes. Outcomes post AKI in vivo that were obtained with the immediate post-injury intraparenchymal injection of 1×10^6 ASCs in CS-IGF-1 were compared with those seen with PBS only, hydrogel injection only, ASCs only, ASCs with unmodified hydrogel, and sham animals. Injecting ASCs together with the IGF-1–modified chitosan hydrogel into the remaining kidney (post unilateral nephrectomy) of a mouse with ischemia–reperfusion injury AKI resulted in prolonged ASC survival in the kidney, increased cell proliferation, decreased CD68–positive macrophage numbers, and TNFα levels, as well as reduced apoptotic indices in the kidney. In parallel, numerous proangiogenic genes were upregulated in the kidney, and microvascular densities were increased, while injury scores were significantly reduced. Return of renal function was better although never reached baseline levels by day 28. Importantly, and as previously reported, extracellular matrix deposition and interstitial fibrosis, and respective gene expression profiles were most effectively improved by ASCs embedded in IGF-1–modified hydrogel.

Because it has been shown previously that both IGF-1 therapy in experimental AKI improves outcomes and that IGF-1 expression by MSCs is critical to their renoprotective activity in AKI, the observations by Feng et al. are per se not unexpected. What is a novel observation in these studies is the fact that ex vivo exposure of IGF-1 receptor-expressing and IGF-1–secreting ASCs to a hydrogel that can activate IGF-1 receptors boosts the expression of IGF-1, HGF, EGF, and proangiogenic factors in these cells. This pattern of increased IGF-1 expression by ASCs to the IGF-1 C domain-modified hydrogel does not follow the canonical interaction between this ligand and its tyrosine kinase–linked receptor, i.e., where increased stimulation of such receptors by their ligand results in their downregulation and vice versa. The signaling and molecular mechanisms that underlie such a response remain to be elucidated.

In AKI, SDF-1 (CXCL12) is promptly and significantly upregulated in the kidney, a response that resembles the physiology of SDF-1 in the stem cell niches of the bone marrow. Specifically, when hematopoietic stem cells are infused intravenously to a patient who is undergoing a bone marrow transplant post myeloablution, SDF-1 levels in the bone marrow niches are sufficient to recruit these CXCR4-expressing (SDF-1 receptor) hematopoietic stem cells to the bone marrow where they engraft and differentiate. In the kidney post acute injury, a “facultative stem cell niche” is temporarily expressed that mediates the augmented recruitment of administered MSCs through their expression of CXCR4, the cognate receptor for this chemokine. However, MSCs that home to the injured kidney do not engraft or differentiate, but instead temporarily exert local anti-inflammatory and trophic actions before they leave the kidney and undergo apoptosis. It would, therefore, be of substantial interest to assess whether the treatment with ASCs in the IGF-1–modified hydrogel alters the tubular and/or ASC expression of SDF-1, a well-recognized and potent survival factor.

The authors, likely for practical reasons, decided to test their ASC plus IGF-1–modified hydrogel therapy in a single kidney model of AKI. This study design immediately illustrates one of the principal limitations of this therapy, i.e., the need for bilateral injections because the vast majority of patients have AKI that obviously affects both of their kidneys. Bilateral intraparenchymal injections are invasive and may have procedure-related complications. As discussed above, the timing of such therapy must be very early after a renal insult, and this depends on the use of diagnostic biomarkers and not on a delayed rise in serum creatinine levels, a retrospective, non-specific marker of a decline in kidney function. It would be conceivable to consider such therapy as suitable for patients with only one kidney, e.g., kidney transplants or those who are known to have or develop AKI following uninephrectomy for a number of indications. Finally, because in successful preclinical and clinical studies, MSCs were either administered into the suprarenal aorta, intravenously or intraperitoneally, it would be of value to see a comparison of the present study’s results with those that are seen with an intravascular route of administration, which is less invasive and targets both kidneys. Similarly, potentiation of renoprotection by ASCs or MSCs might be accomplished through in vitro preconditioning with IGF-1 alone, which has been previously shown to be beneficial, or culturing them on the IGF-1–modified hydrogel, both of which may actually prove to be a better approach.

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DISCLOSURES

None.

REFERENCES

Parathyroidectomy or Calcimimetic to Treat Hypercalcemia after Kidney Transplantation?

Masafumi Fukagawa* and Tilman B. Drüeke†

*Division of Nephrology, Endocrinology, and Metabolism, Tokai University School of Medicine, Isehara, Japan; and †Institut National de la Santé et de la Recherche Médicale Unit 1018, Team 5, Centre de Recherche en Épidémiologie et Santé des Populations, Hôpital Paul Brousse, Paris-Sud University and Versailles Saint-Quentin-en-Yvelines University (Paris-Ile-de-France-Ouest University), Villejuif, France


Secondary hyperparathyroidism is a nearly constant feature of CKD. Other than its well known effects on the bone, namely osteitis fibrosa and mixed bone disease, it has been incriminated in many other complications of patients with CKD: vascular and other soft tissue calcification, cardiovascular morbidity and mortality, hematologic disorders, neurologic disturbances, and endocrine abnormalities. If insufficiently treated or left untreated, severe parathyroid hyperplasia will ensue in many patients with ESRD, ultimately leading to clonal, tumoral growth.1 The treatment of severe hyperparathyroidism in patients with ESRD by either parathyroidectomy or cinacalcet generally leads to a marked decrease in serum calcium, phosphate, parathyroid hormone (PTH), and as shown more recently, serum fibroblast growth factor-23.2,3

When such patients receive a kidney graft, generally polyclonal parathyroid hyperplasia will slowly regress because of the recovery of renal function and correction of metabolic and endocrine disturbances. This is not so with tumoral growth, where the response of parathyroid secretion and proliferation to physiologic regulatory mechanisms is reduced or lost.4,5 The