Adult stem cells as a tool for kidney regeneration

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Abstract

Kidney regeneration is a challenging but promising strategy aimed at reducing the progression to end-stage renal disease (ESRD) and improving the quality of life of patients with ESRD. Adult stem cells are multipotent stem cells that reside in various tissues, such as bone marrow and adipose tissue. Although intensive studies to isolate kidney stem/progenitor cells from the adult kidney have been performed, it remains controversial whether stem/progenitor cells actually exist in the mammalian adult kidney. The efficacy of mesenchymal stem cells (MSCs) in the recovery of kidney function has been demonstrated in animal nephropathy models, such as acute tubular injury, glomerulonephritis, renal artery stenosis, and remnant kidney. However, their beneficial effects seem to be mediated largely via their paracrine effects rather than their direct differentiation into renal parenchymal cells. MSCs not only secrete bioactive molecules directly into the circulation, but they also release various molecules, such as proteins, mRNA, and microRNA, in membrane-covered vesicles. A detailed analysis of these molecules and an exploration of the optimal combination of these molecules will enable the treatment of patients with kidney disease without using stem cells. Another option for the treatment of patients with kidney disease using adult somatic cells is a direct/indirect reprogramming of adult somatic cells into kidney stem/progenitor cells. Although many hurdles still need to be overcome, this strategy will enable bona fide kidney regeneration rather than kidney repair using remnant renal parenchymal cells.

Key words: Adult stem cells; Direct reprogramming; Extracellular vesicles; Mesenchymal stem cells; Paracrine factors; Indirect reprogramming

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Core tip: Although intensive studies have been performed to isolate kidney stem/progenitor cells from the mammalian adult kidney, whether stem/progenitor cells actually exist in the adult kidney is still debated. Mesenchymal stem cells seem to exert beneficial effects via paracrine effects rather than by direct differentiation into renal parenchymal cells. In this review, we also introduce potential roles of extracellular vesicles released
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from stem cells and direct/indirect reprogramming of adult somatic cells by which kidney stem/progenitor cells will be formed in the future.

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**INTRODUCTION**

The kidney is a vital organ that plays various roles, such as the excretion of waste products; regulation of systemic fluid volume, electrolytes, and pH; maintenance of systemic blood pressure; and erythropoietin production. These functions are performed by the nephrons, the functional units of the kidney. If the structure and/or function of the nephrons are damaged because of diseases, such as diabetes, hypertension, and glomerulonephritis, and if such damages continue to progress, renal function gradually deteriorates. The kidney finally becomes unable to perform its critical roles, resulting in renal failure.

There are two therapeutic options for the treatment of end-stage renal disease (ESRD). One is dialysis therapy, which compromises patients’ quality of life and cannot substitute for all kidney functions. Another is renal transplantation, which is limited because of the lack of sufficient donors. To explore a better treatment for ESRD, it is necessary to find strategies to regenerate the kidney. In this respect, stem cell therapy for the kidney has been intensively studied recently.

Stem cells are defined as cells that are capable of self-renewal and can differentiate into a variety of phenotypes. Adult stem cells (ASCs) are multipotent stem cells that reside in various tissues, such as the bone marrow, adipose tissue, and skeletal muscle. In this article, we review the possibility of kidney regeneration using ASCs.

**Stem cells in the embryonic kidney**

Although stem/progenitor cells in the embryonic kidney are beyond the scope of this review, we briefly describe the process of kidney organogenesis, because genetic programs that are activated during kidney organogenesis are reactivated in disease states, such as acute tubular injuries. Kidney organogenesis initiates with the interaction of the ureteric bud (UB) derived from the Wolffian duct with the metanephric mesenchyme (MM). A proportion of the MM is located adjacent to the UB, called the capsular mesenchyme (CM). The CM then aggregates at the tip of the UB and differentiates into all epithelial cells of nephrons, except the collecting tubules. It is now well established that the CM contains stem/progenitor cells for kidney organogenesis. The CM expresses unique transcription factors, such as *Pax2*, *Six2*, and *Sall1*. Although stem cells in the embryonic kidney are a promising source for kidney regeneration, their clinical use is strictly limited mainly because of the ethical concerns and small number of stem cells. Therefore, the search for stem/progenitor cells in the adult kidney has been intensively performed.

**Stem/progenitor cells in the adult kidney**

Four different methods have been used in an attempt to isolate stem/progenitor cells from the adult kidney (Table 1).

**Label-retaining cells (LRCs):** To conserve the proliferation capacity for a lifetime and to prevent genetic injuries during mitosis, stem cells cycle very slowly. Stem/progenitor cells in the kidney were isolated using this property. Cells were pulse-labeled with a dye, such as 5-bromo-2-deoxyuridine. Then, slow-cycling LRCs were detected following a chase period. Maeshima et al detected LRCs predominantly in the renal tubular cells of the adult rat kidney. LRCs proliferated in response to ischemia/reperfusion injury and contributed to the repair of renal tubules. In another study, Maeshima et al also demonstrated that LRCs were integrated into epithelial components of the nephron when transplanted into the metanephric kidney, suggesting that LRCs were multipotent stem cells. Oliver et al detected LRCs in the kidney papilla of adult rats. LRCs proliferated after the induction of ischemia in the kidney and migrated toward the medulla. They also injected renal papillary cells into the subcapsular area of the kidney and found that some cells were incorporated into renal tubules.

**Side population cells:** Because stem cells extrude dyes, such as Rhodamine 123 and Hoechst 33342, via the ATP-binding cassette protein, they are located in a unique position on the fluorescent-assisted cell sorting scattered plot and are called side population (SP) cells. Iwatani et al isolated SP cells from the adult rat kidney. However, the cells did not participate in the kidney repair following experimental glomerulonephritis or gentamicin-induced nephropathy. Hishikawa et al isolated SP cells from the adult murine kidney. These cells expressed muscin/MyoR and improved renal function when injected systemically with the induction of acute tubular injury by cisplatin administration. Furthermore, SP cells expressed renal-protective factors, such as hepatocyte growth factor (HGF), vascular endothelial growth factor, and bone morphogenetic protein 7 in a cisplatin-induced acute kidney injury (AKI) model. Challen et al also isolated SP cells from the adult murine kidney. These cells were located predominantly in the proximal tubules and integrated into the MM- and UB-derived structures when injected into the embryonic kidney, suggesting that they were multipotent stem cells. However, these cells were barely incorporated into the renal tissues when...
administered to an adriamycin-induced kidney injury model, although renal function was recovered, probably because of their paracrine effect.

**Cell surface markers:** Cell surface markers, such as CD133, were used to isolate stem/progenitor cells from the adult kidney. Although CD133 is not a specific marker for stem cells, it is a universal marker for stem cells in other tissues, such as hematopoietic stem cells, vascular endothelial progenitor cells (EPCs), and cancer stem cells\[13,14\]. Bussolati et al\[15\] isolated CD133+ cells from human renal cells. These cells expressed Pax2, but not CD34 or CD45, markers for hematopoietic stem cells. They could also be induced to differentiate into tubular epithelial cells and endothelial cells in vitro. When these cells were administrated intravenously in a glycerol-induced AKI model of severe combined immune deficiency (SCID) mice, they were incorporated predominantly into the proximal and distal tubules. Sagrinati et al\[16\] isolated CD133+ and CD24+ cells from human parietal epithelial cells in the Bowman’s capsule after culturing glomeruli. When cultured in appropriate conditions, these cells were differentiated into tubular epithelial cells, osteogenic cells, adipocytes, and neuronal cells. These cells were integrated predominantly into renal tubules when they were injected intravenously in SCID mice that were treated with glycerol to induce acute tubular injuries. Dekel et al\[17\] isolated stem cell antigen-1 (Sca1)-positive cells from the adult mouse kidney. The Sca1+ cells were located mainly in the papilla. When these cells were administrated to an ischemia-induced AKI model, some of these cells were integrated into renal tubules.

**Cell culture:** A unique cell population was isolated during the culture of dispersed cells derived from the adult kidney. Gupta et al\[18\] isolated progenitor-like cells from the adult rat kidney that express vimentin, CD90, Pax2, and Oct4, a marker for embryonic stem cells. These cells were incorporated into renal tubules when injected under the capsule of the kidney or intravenously, following ischemia-reperfusion injury of the kidney.

### Arguments against the presence of stem/progenitor cells in the mammalian adult kidney

Although aforementioned results suggest that renal stem/progenitor cells exist in the adult kidney, some reports demonstrated that differentiated renal tubular cells, but not renal stem/progenitor cells, can completely regenerate renal tubules after injury. Humphreys et al\[19\] used genetic fate-mapping techniques in which renal epithelial cells derived from the CM (from the Bowman’s capsule to the junction of the connecting segment and collecting duct) were labeled with either β-galactosidase or red fluorescent protein (RFP). After ischemia-reperfusion injury, approximately 50% of the proximal tubular cells coexpressed both RFP and Ki67, a cell proliferation marker that is expressed during the S-M phases of the cell cycle. These findings suggested that intrinsic renal tubular cells proliferate in response to injury. Furthermore, approximately 95% of tubular epithelial cells expressed RFP prior to injury, after one cycle of injury, and after two cycles of injury, indicating that no dilution of the RFP+ tubular epithelial cells occurred. These results suggested that differentiated renal epithelial cells proliferate well in response to the injury and that stem and/or progenitor cells residing in the interstitium did not participate in the regeneration of the tubules. Kusaba et al\[20\] used a genetically modified mouse in which the tdTomato protein, which fluoresces in a red color, expressed only in differentiated proximal renal tubules. No dilution of tdTomato+ cells was observed after ischemia-reperfusion injury, suggesting that stem/progenitor cells in renal tubules did not participate in the regeneration of renal tubules. Furthermore, they observed that tdTomato+ proximal tubules expressed CD24 and CD133, markers for stem/progenitor cells. These findings suggested that renal tubules were dedifferentiated and expressed stem cell markers during their proliferation and participation in the repair of renal tubules. These results did not support that stem/progenitor cells in renal tubules and in the interstitium participated in the regeneration of renal tubules.

| Species | Isolation method | Stem cell markers | Location | Incorporation into kidney tubules | Ref. |
|---------|------------------|-------------------|----------|----------------------------------|------|
| Rat     | Label retaining  | Sca-1, CD45       | Proximal tubule | Yes | Maeshima et al\[8\] |
| Rat     | Label retaining  | CD34              | Papilla   | Yes | Oliver et al\[9\] |
| Rat     | Side population  | Sca-1, CD45       | Some were derived from bone | No | Hishikawa et al\[10\] |
| Mouse   | Side population  | Sca-1             | Intersitial | NE | Challen et al\[11\] |
| Human   | Marker           | CD133             | Proximal tubule | Yes (rare) | Bussolati et al\[12\] |
| Human   | Marker           | CD133, CD24       | Parietal epithelium in the Bowman’s capsule | Yes | Sagrinati et al\[13\] |
| Mouse   | Marker           | Sca-1             | Papilla   | Yes | Dekel et al\[16\] |
| Rat     | Culture          | Pax2, Oct4        | Proximal tubule | Yes | Gupta et al\[17\] |

NE: Not examined.
Many studies have demonstrated the efficacy of BMMSCs in the treatment of kidney disease using animal models of AKI\(^22\)–\(^26\), podocyte injury\(^27\), glomerulonephropathy\(^28\)–\(^30\), a remnant kidney\(^31\), and kidney transplantation\(^32\) (Table 2). Although early studies indicated BMMSCs could be differentiated into renal epithelial cells\(^22,23\) and mesangial cells\(^30\), recent evidence suggests that the differentiation capacity of BMMSCs is limited. Thus, BMMSCs do not appear to differentiate into renal parenchymal cells \textit{in vivo}\(^33\). The beneficial effects of BMMSCs on renal function seem to be largely mediated by paracrine factors produced by BMMSCs that have anti-apoptotic, proangiogenic, and/or immune modulatory effects\(^34\)–\(^36\). The transdifferentiation of BMMSCs observed in early reports may reflect cell fusion. If BMMSCs fuse with resident cells in the kidney, they acquire a phenotype of resident cells. Thus, it appears as if BMMSCs were differentiated into resident cells. Indeed, several reports have demonstrated that BMMSCs are capable of fusing with other cell types\(^37,38\). A phase I clinical study evaluated the safety and efficacy of allogenic BMMSC administration for the prevention of AKI after open-heart surgery\(^39\). This study enrolled 16 patients who required on-pump cardiac surgery and who were at a high risk of postoperative AKI due to underlying chronic kidney disease (CKD), advanced age, diabetes mellitus, and congestive heart failure. Allogenic BMMSCs were injected into the suprarenal aorta after surgery. The primary objective was the safety of BMMSC administration. The secondary objective was the efficacy of this treatment compared with well-matched historical controls. This treatment appeared to be both safe and effective as no adverse events related to the procedure were reported, and renal function was well preserved post-operatively, with no patients requiring hemodialysis after surgery, whereas 20% of the controls developed AKI. This is the only clinical trial published so far in which ASCs were used to treat kidney disease.  

**ADSCs:** ADSCs are another type of MSCs residing...
in subcutaneous adipose tissues. Because the subcutaneous adipose tissues are abundant and can be easily harvested using liposuction, ADSCs are promising stem cells for clinical use. The efficacy of ADSC administration in the treatment of kidney disease has been demonstrated in animal models of AKI, glomerulonephropathy, renal artery stenosis, and progressive renal fibrosis (Table 3). ADSCs also seem to recover renal function largely via paracrine effects.

**EPCs**: EPCs were originally isolated from human peripheral blood using CD34 as a marker for positive selection. The CD34+ mononuclear blood cells obtained the characteristics of vascular endothelial cells (VECs) when cultured on fibronectin-coated dishes. EPCs were reportedly incorporated in ischemic tissues in vivo and expressed markers for VECs such as CD31 when introduced into the circulation using a hindlimb ischemia model. The efficacy of EPC administration in the recovery of renal function was reported in animal models of AKI and renal artery stenosis. Interestingly, the function of EPCs was deteriorated in CKD patients, suggesting that the autologous transplantation of EPCs may not be suitable for the treatment of CKD.

**Umbilical cord blood-derived MSCs**

Umbilical cord blood (UCB) contains MSCs, and the efficacy of UCB administration in the restoration of renal function has been reported in animal AKI models. Morigi et al. injected human UCB-derived MSCs to immunodeficient mice with cisplatin-induced acute tubular injury. They demonstrated that these cells ameliorated tubular injury, resulting in the recovery of renal function. They also cocultured UCB-derived MSCs with cisplatin-treated proximal tubular cells (HK-2 cells) and demonstrated that the expression of HGF was particularly induced and that of interleukin 1-β and tumor necrosis factor-α was significantly decreased in the coculture system. These findings suggested that the modulation of paracrine factors in the kidney was implicated in the UCB-induced recovery of renal function. Panepucci et al. compared the gene expression profile of UCB-derived MSCs and BM-MSCs. Although both MSCs expressed similar sets of genes, BM-MSCs predominantly expressed a set of genes related to antimicrobial activity and osteogenesis, whereas UCB-derived MSCs predominantly expressed genes related to matrix remodeling and angiogenesis, suggesting that UCB-derived MSCs and BM-MSCs may have distinct activities in vivo.

**Amniotic fluid stem cells**

Human amniotic fluid contains stem cells derived from embryos, and thus, is a promising source of stem cells. The efficacy of human amniotic fluid stem cells (HAFSCs) has been demonstrated in animal models of AKI. Houser et al. compared the characteristics of HAFSCs with that of BM-MSCs. They found that compared with BM-MSCs, HAFSCs had a more potent anti-apoptotic activity against renal tubular cells but lesser stimulatory activity for the proliferation of renal tubular cells. They also demonstrated that HAFSCs and BM-MSCs expressed distinct sets of paracrine factors, suggesting that HAFSCs and BM-MSCs may have distinct activities in vivo.

**Direct/indirect reprogramming of adult somatic cells**

Another strategy for kidney regeneration is to create...
pluripotent stem cells and progenitor cells, whose destination is limited to one or several cell lineages, or terminally differentiated renal parenchymal cells from adult somatic cells via direct/indirect reprogramming. Indirect reprogramming is a strategy in which adult somatic cells are induced to dedifferentiate into pluripotent stem cells and re-differentiate into specific cell types. Direct reprogramming involves a strategy in which adult somatic cells are induced to differentiate directly into another cell type. Since the discovery of induced pluripotent stem (iPS) cells, it is not difficult to prepare iPS cells from various adult somatic cells. Indeed, several reports have demonstrated a successful preparation of nephrogenic intermediate mesoderm from which the MM and the UB derive, using iPS cells. However, several hurdles remain to be overcome before iPS cells can be used practically to regenerate the kidney. First, the efficiency of iPS cell preparation from adult somatic cells is still low. Second, it is still challenging to prepare kidney stem/progenitor cells that exist in the CM from iPS cells. Third, even if kidney stem/progenitor cells are successfully created from iPS cells, it is difficult to continue to culture and expand those stem/progenitor cells while maintaining their unique properties. The reason for this limitation is that the niche for kidney stem/progenitor cells has not been clearly understood. Therefore, further studies will be required to use indirect reprogramming for kidney regeneration. Recently, several reports have demonstrated that a direct reprogramming method was useful for kidney regeneration. Hendry et al. introduced 6 transcription factors into a human adult renal proximal tubular cell line. These cells were localized in Six2+ and Wilm’s tumor 1 + compartment in an ex vivo organoid culture assay. These findings suggest that these cells obtained properties similar to kidney stem/progenitor cells. Papadimou et al. incubated permeabilized human BM-MSCs with the extracts of human proximal tubular epithelial cells and obtained a cell population similar to proximal renal tubular cells. These cells were integrated in tubular structures in an ex vivo organoid culture assay. Furthermore, these cells were engrafted in renal tubules when administered to a cisplatin-induced AKI model. Therefore, direct reprogramming seems to be a promising strategy for kidney regeneration. It has been reported that iPS cells derived from various cell types are not identical in their differentiation capacity, probably because iPS cells maintain epigenetic memory of their parental cells. Thus, it may be better to use renal parenchymal cells for reprogramming than cells derived from tissues other than the kidney.

**Possible roles of extracellular vesicles released from MSCs**

MSCs not only secrete bioactive molecules directly into the circulation but also release extracellular vesicles (EVs), such as exosomes that contain proteins, mRNA, and microRNA. Several reports have demonstrated that EV administration restored the kidney function in animal models of AKI. Bruno et al. isolated EVs from supernatants of human BM-MSC and examined their effects on the proliferation and apoptosis in renal epithelial cells. EVs were incorporated in renal epithelial cells in vitro and the incorporation depended on CD44 and β1-interin. EVs stimulated the proliferation and inhibited apoptosis of renal epithelial cells. These effects were diminished when EVs were treated with RNase prior to administration. Furthermore, the authors administered EVs to immunodeficient mice with glycerol-induced acute tubular injury and demonstrated that EV administration restored renal function. Moreover, these beneficial effects were diminished when EVs were pretreated with RNase. Tomasoni et al. isolated EVs from supernatants of human BM-MSC and demonstrated that EVs contained mRNA for the insulin-like growth factor-1 receptor (IGF1R). When cisplatin-treated renal epithelial cells were incubated with EVs, proliferative capacity of renal epithelial cells increased significantly; however, the stimulatory effect was diminished when the expression of IGF1R mRNA in BM-MSCs was suppressed using small interfering RNA to IGF1R prior to EV harvest. Zhou et al. administered EVs harvested from human UCB-derived MSCs to a rat model of cisplatin-induced AKI. EV administration significantly restored renal function and morphology. Therefore, EVs seem to contain various bioactive molecules that can be used for the treatment of kidney injury.

**FUTURE DIRECTIONS**

It seems that there are two major directions to improve the quality of stem cell therapy for kidney diseases. One is to analyze bioactive molecules released from stem cells in more details. If an ideal combination of bioactive proteins, mRNA, and/or microRNA is elucidated, stem cells per se will not be necessary in the future. Although BM-MSCs have been used in the clinical setting to treat patients with cardiovascular disease, concern about tumorigenesis still remains. Furthermore, BM-MSCs isolated under uremic conditions have less capacity for the proliferation, survival, and secretion of paracrine factors compared with those isolated from normal controls. Patients who need stem cell therapy are probably not a suitable source of high-quality stem cells, indicating that autologous stem cell transplantation may not be effective in these patients. Therefore, cell-free therapy seems attractive. Another alternative is to explore strategies to directly and/or indirectly reprogram adult somatic cells into kidney stem/progenitor cells. If efficient and safe methods to induce direct/indirect reprogramming are explored, bona fide kidney regeneration rather than kidney repair using remnant renal parenchymal cells will be possible in the future.

**CONCLUSION**

Although intensive studies have been performed to isolate kidney stem/progenitor cells from the adult
kidney, it is debated whether kidney stem/progenitor cells actually exist in the adult kidney. There are no specific markers and/or assays to discriminate kidney stem/progenitor cells from dedifferentiated renal epithelial cells. MSCs are effective to repair the kidney in various animal models; however, their beneficial effects can be largely attributed to paracrine factors that are secreted from MSCs. In addition, MSCs release EVs that contain mRNA and microRNA as well as proteins. These EV-derived molecules may also play beneficial roles in the repair of the kidney. It will be necessary to elucidate ideal combinations of the molecules released from MSCs to establish a strategy for the maximal stimulation of kidney repair without using stem cells. It will also be necessary to explore strategies to directly and/or indirectly reprogram somatic cells to kidney stem/progenitor cells to regenerate the kidney.

REFERENCES
1. Morrison SJ, Shah NM, Anderson DJ. Regulatory mechanisms in stem cell biology. Cell 1997; 88: 287-298 [PMID: 9039255]
2. Jiang Y, Vaessen B, Lenvik T, Blackstad M, Reyes M, Verfaillie CM. Multipotent progenitor cells can be isolated from postnatal murine bone marrow, muscle, and brain. Exp Hematol 2002; 30: 896-904 [PMID: 12160841]
3. Schäffer A, Büchler C. Concise review: adipose tissue-derived stromal cells--basic and clinical implications for novel cell-based therapies. Stem Cells 2007; 25: 818-827 [PMID: 17420225 DOI: 10.1634/stemcells.2006-0589]
4. Kobayashi A, Valerius MT, Mugford JW, Carroll TJ, Self M, Oliver SG, McMahon AP. Six2 defines and regulates a multipotent self-renewing nephron progenitor population throughout mammalian kidney development. Cell Stem Cell 2008; 3: 169-181 [PMID: 18682239 DOI: 10.1016/j.stem.2008.05.020]
5. Reidy KJ, Rosenblum ND. Cell and molecular biology of kidney development. Semin Nephrol 2009; 29: 321-337 [PMID: 19615554 DOI: 10.1016/j.semnephrol.2009.03.009]
6. Maeshima A, Yamashita S, Nogima J. Identification of renal progenitor-like tubular cells that participate in the regeneration processes of the kidney. J Am Soc Nephrol 2003; 14: 3138-3146 [PMID: 14638912]
7. Maeshima A, Sakurai H, Nigam SK. Adult kidney tubular cell population showing phenotypic plasticity, tubulogenic capacity, and integration capability into developing kidney. J Am Soc Nephrol 2006; 17: 2443-2456 [PMID: 16885410 DOI: 10.1681/ASN.2006010089]
8. Gupta S, Verfaillie C, Chimeleswic D, Kren S, Eidman K, Connaire J, Heremans Y, Lund T, Blackstad M, Jiang Y, Luttan A, Rosenberg ME. Isolation and characterization of kidney-derived stem cells. J Am Soc Nephrol 2006; 17: 3306-3314 [PMID: 17093069 DOI: 10.1681/ASN.2005020195]
9. Humphreys BD, Valerius MT, Kobayashi A, Mugford JW, Soeung S, Duffield JS, McMahon AP, Bonventre JV. Intrinsically epithelial cells repair the kidney after injury. Cell Stem Cell 2008; 2: 284-291 [PMID: 18371453 DOI: 10.1016/j.stem.2008.01.014]
10. Kusaba T, Lalii M, Kramann R, Kobayashi A, Humphreys BD. Differentiated kidney epithelial cells repair injured proximal tubule. Proc Natl Acad Sci USA 2014; 111: 1527-1532 [PMID: 24125783 DOI: 10.1073/pnas.1310535110]
11. Ronconi E, Maes A, Angiulli ML, Lazzeri E, Mazzinghi B, Ballerini L, Parente E, Becherucci F, Gauci M, Carini M, Maggi E, Serio M, Gatti LG, Lasagni L, Romagnani S, Romagnani P. Regeneration of glomerular podocytes by human renal progenitors. J Am Soc Nephrol 2009; 20: 322-332 [PMID: 19092120 DOI: 10.1681/ASN.2008070709]
12. Herrera MB, Bussolati B, Bruno S, Fonsato V, Romanazzi GM, Camussi G. Mesenchymal stem cells contribute to the repair of adult acinar epithelial injury. Int J Mol Med 2004; 14: 1035-1041 [PMID: 15547670]
13. Morigi M, Imeriti B, Zioja C, Carnia D, Pisoni S, Abbate M, Rottoli D, Angelotti ML, Lazzari E, Mazzinghi B, Ballerini L, Parente E, Becherucci F, Gauci M, Carini M, Maggi E, Serio M, Vannelli GB, Lasagni L, Romagnani S, Romagnani P. Regeneration of glomerular podocytes by human renal progenitors. J Am Soc Nephrol 2004; 15: 1794-1804 [PMID: 15213267]
14. Lange C, Tögel F, Ittrich H, Clayton F, Nolte-Ernsting C, Zander AR, Westenfelder C. Administered mesenchymal stem cells enhance recovery from ischemia/reperfusion-induced acute renal failure in rats. Kidney Int 2005; 68: 1613-1617 [PMID: 16146388 DOI: 10.1111/j.1523-1755.2005.00573.x]
15. Morizzi M, Intra M, Imeriti B, Carnia D, Abbate M, Rota C, Rottoli D, Benigni A, Perico N, Zioja C, Rambaldi A, Remuzzi A, Remuzzi G. Human bone marrow mesenchymal stem cells accelerate recovery of acute renal injury and prolong survival in mice. Stem Cells 2008; 26: 2075-2082 [PMID: 18499895 DOI: 10.1634/stemcells.2007-0795]
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26 Reis LA, Borges FT, Simões MJ, Borges AA, Sinigaglia-Coimbra R, Schor N. Bone marrow-derived mesenchymal stem cells repaired bone but did not prevent gentamicin-induced acute kidney injury through paracrine effects in rats. PLoS One 2012; 7: e44092 [PMID: 22807165 DOI: 10.1371/journal.pone.0044092]

27 Zoja C, Garcia PB, Rota C, Conti S, Gagliardini E, Corna D, Zanchi C, Bigini P, Benigni A, Remuzzi G, Morigi M. Mesenchymal stem cell therapy promotes renal repair by limiting glomerular podocyte and progenitor cell dysfunction in adriamycin-induced nephropathy. Am J Physiol Renal Physiol 2012; 303: F1370-F1381 [PMID: 22952284 DOI: 10.1152/ajprenal.00507.2012]

28 Kunter U, Rong S, Djuric Z, Boor P, Müller-Newen G, Ya D, Floege J. Transplanted mesenchymal stem cells accelerate glomerular healing in experimental glomerulonephritis. J Am Soc Nephrol 2006; 17: 2202-2212 [PMID: 16795013 DOI: 10.1681/ASN.2005080815]

29 Ninchuk V, Gross O, Segerer S, Hoffmann R, Radomska E, Park KM, Hsiao LL, Kelley VR, Scadden DT, Cheong SK, Mok PL, Leong CF. Differentiation of adult mesenchymal stem cells: a new therapeutic tool for AKI. DOI: 10.1038/nrneph.2009.229]

30 Chen YT, Sun CK, Lin YC, Chang LT, Chen YL, Tsai TH, Chung SY, Chua S, Kao YH, Yen CH, Shao PL, Chung KC, Leu S, Yip HK. Adipose-derived mesenchymal stem cell protects kidneys against ischemia-reperfusion injury through suppressing oxidative stress and inflammatory reaction. J Transl Med 2011; 9: 51 [PMID: 21545725 DOI: 10.1186/1479-5876-9-51]

31 Katsuno T, Ozaki T, Saka Y, Furushashi K, Kim H, Yasuda K, Yamamoto T, Sato W, Tsuboi N, Mizuno M, Ito Y, Imai E, Matsu S, Maruyama S. Low serum cultured adipose tissue-derived stromal cells ameliorate acute kidney injury in rats. Cell Transplant 2013; 22: 287-297 [PMID: 22963874 DOI: 10.3727/096369812X665019]

32 Furushashi K, Tsuboi N, Shimizu A, Katsuno T, Kim H, Saka Y, Ozaki T, Sado Y, Imai E, Matsu S, Maruyama S. Serum-starved adipose-derived stromal cells ameliorate crescentic GN by promoting immunoregulatory macrophages. J Am Soc Nephrol 2013; 24: 587-603 [PMID: 23471196 DOI: 10.1681/ASNA.2012032064]

33 Eirin A, Zhu XY, Krier JD, Tang H, Jordan KL, Grande JP, Lerman A, Textor SC, Lerman LO. Adipose tissue-derived mesenchymal stem cells improve revascularization outcomes to restore renal function in swine atherosclerotic renal artery stenosis. Stem Cells 2012; 30: 1030-1041 [PMID: 22298382 DOI: 10.1002/stem.1047]

34 Zhu XY, Urbietta-Caceres V, Krier JD, Textor SC, Lerman A, Lerman LO. Mesenchymal stem cells and endothelial progenitor cells decrease renal injury in experimental swine renal artery stenosis through different mechanisms. Stem Cells 2013; 31: 117-125 [PMID: 23097349 DOI: 10.1002/stem.1263]

35 Ebrahimi B, Eirin A, Li Z, Zhu XY, Zhang X, Lerman A, Textor SC, Lerman LO. Mesenchymal stem cells improve medullary inflammation and fibrosis after revascularization of swine atherosclerotic renal artery stenosis. Stem Cells 2013; 31: 1215-1223 [PMID: 23844047 DOI: 10.1002/stem.2134]

36 Donizetti-Oliveira C, Semedo P, Burgos-Silva M, Cenedeze MA, Malheiro DM, Reis MA, Pacheco-Silva A, Câmara NO. Adipose tissue–derived stem cell treatment prevents renal disease progression. Cell Transplant 2012; 21: 1727-1741 [PMID: 22530561 DOI: 10.3727/096369812X623925]

37 Ashara T, Murohara T, Sullivan A, Silver M, van der Zee R, Li T, Wittenbach B, Schattenmann G, Isner JM. Activation of putative progenitor endothelial cells for angiogenesis. Science 1997; 275: 964-967 [PMID: 920076]

38 Patschan D, Krupincza P, Patschan S, Zhang Z, Hamby C, Goligorsky MS. Dynamics of mobilization and homing of endothelial progenitor cells after acute renal ischemia: modulation by ischemic preconditioning. Am J Physiol Renal Physiol 2006; 291: F176-F185 [PMID: 16478972 DOI: 10.1152/ajprenal.00454.2005]

39 Chade AR, Zhu X, Lavi R, Krier JD, Pilsaru S, Simari RD, Napoli C, Lerman A, Lerman LO. Endothelial progenitor cells restore renal function in chronic experimental renovascular disease. Circulation 2009; 119: 547-557 [PMID: 19153272 DOI: 10.1161/CIRCULATIONAHA.108.788653]

40 Chade AR, Zhu XY, Krier JD, Jordan KL, Textor SC, Grande JP, Lerman A, Lerman LO. Endothelial progenitor cells homing and renal repair in experimental renovascular disease. Stem Cells 2010; 28: 1059-1047 [PMID: 20506499 DOI: 10.1002/stem.426]

41 Jie KE, Zukova MA, Bergevoet MW, Westerweel PE, Rasmussen M, Blankestijn PJ, Boer WH, Beam B, Verhaar MC. Progenitor cells and vascular function are impaired in patients with chronic kidney disease. Nephrol Dial Transplant 2010; 25: 1875-1882 [PMID: 20083473 DOI: 10.1093/ndt/gfp749]

42 Morigi M, Rota C, Montemuro T, Montelatici E, Lo Cicero V, Imberti B, Abbate M, Zoja C, Casisi P, Longaretti L, Rebulla P, Introna M, Capelli C, Benigni A, Remuzzi G, Lazzari L. Life-sparing effect of human cord blood-mesenchymal stem cells in...
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experimental acute kidney injury. Stem Cells 2010; 28: 513-522 [PMID: 20049901 DOI: 10.1002/stem.293]

Shalaby RH, Rashid LA, Ismaail AE, Madkour NK, Elwakeel SH. Hematopoietic stem cells derived from human umbilical cord blood ameliorate cisplatin-induced acute renal failure in rats. Am J Stem Cells 2014; 3: 83-96 [PMID: 25232508]

Panpepuci RA, Siufi JL, Silva WA, Proto-Siquiera R, Neder L, Orellana M, Rocha V, Covas DT, Zago MA. Comparison of gene expression of umbilical cord vein and bone marrow-derived mesenchymal stem cells. Stem Cells 2004; 22: 1263-1278 [PMID: 15579645 DOI: 10.1634/stemcells.2004-0024]

Hauser PV, De Fazio R, Bruno S, Sidi S, Grange C, Bussolati B, Benedetto C, Camussi G. Stem cells derived from human amniotic fluid contribute to acute kidney injury recovery. Am J Pathol 2010; 177: 2011-2021 [PMID: 20724594 DOI: 10.2353/ajpath.2010.091245]

Rota C, Imberti B, Pozzobon M, Piccoli M, De Coppi P, Atala A, Gagliardini E, Xinaris C, Benedetti V, Fabricio AS, Squarcina E, Abbate M, Benigni A, Remuzzi G, Morigi M. Human amniotic fluid stem cell preconditioning improves their regenerative potential. Stem Cells Dev 2012; 21: 1911-1925 [PMID: 22606606 DOI: 10.1089/scd.2011.0333]

Takahashi K, Yamanaka S. Induction of pluripotent stem cells from mouse embryonic and adult fibroblast cultures by defined factors. Cell 2006; 126: 663-676 [PMID: 16904174 DOI: 10.1016/j.cell.2006.07.024]

Takahashi K, Tanabe K, Oshuji M, Narita M, Ichisaka T, Tomoda K, Yamanaka S. Induction of pluripotent stem cells from adult human fibroblasts by defined factors. Cell 2007; 131: 861-872 [PMID: 18035408 DOI: 10.1016/j.cell.2007.11.019]

Araoka T, Mae S, Kurosue Y, Uesugi M, Ohta A, Yamanaka S, Osafune K. Efficient and rapid induction of human iPSCs/ESCs into nephrogenic intermediate mesoderm using small molecule-based differentiation methods. PLoS One 2014; 9: e84881 [PMID: 24454758 DOI: 10.1371/journal.pone.0084881]

Lam AQ, Freedman BS, Mortizane R, Lerou PH, Valerius MT, Bonventre JV. Rapid and efficient differentiation of human pluripotent stem cells into intermediate mesoderm that forms tubules expressing kidney proximal tubular markers. J Am Soc Nephrol 2014; 25: 1211-1225 [PMID: 24357672 DOI: 10.1681/ASN.2013080831]

Mae S, Shono A, Shiota F, Yasuno T, Kajiwara M, Gotoda-Nishimura N, Arai S, Sato-Otubo A, Toyoda T, Takaoka K, Nakayama N. Amniotic mesenchymal stem cells protect against ischemia-reperfusion-induced acute and chronic kidney injury. Nephrol Dial Transplant 2011; 26: 1474-1483 [PMID: 21324974 DOI: 10.1093/ndt/gfr015]

Bruno S, Grange C, Deregibus MC, Calogero RA, Saviozzi S, Collino F, Morando L, Busca A, Falda M, Bussolati B, Tetta C, Camussi G. Mesenchymal stem cell-derived microvesicles protect against acute tubular necrosis. Am J Nephrol 2009; 20: 1053-1067 [PMID: 19389847 DOI: 10.1618/asn.2008070798]

Gatti S, Bruno S, Deregribus MC, Sordi A, Cantaluppi V, Tetta C, Camussi G. Microvesicles derived from human adult mesenchymal stem cells protect against ischaemia-reperfusion-induced acute and chronic kidney injury. Nephrol Dial Transplant 2011; 26: 1474-1483 [PMID: 21324974 DOI: 10.1093/ndt/gfr015]

Bruno S, Grange C, Collino F, Deregibus MC, Cantaluppi V, Biancone L, Tetta C, Camussi G. Microvesicles derived from mesenchymal stem cells enhance survival in a lethal model of acute kidney injury. PLoS One 2012; 7: e33115 [PMID: 22431999 DOI: 10.1371/journal.pone.0033115]

Tomasoni S, Longaretti L, Rota C, Morigi M, Conti S, Gotti E, Capelli C, Intrafoni M, Remuzzi G, Benigni A. Transfer of growth factor receptor mRNA via exosomes unravels the regenerative effect of mesenchymal stem cells. Stem Cells Dev 2013; 22: 772-780 [PMID: 23082760 DOI: 10.1089/scd.2012.0266]

Zhou Y, Xu H, Xu W, Wang B, Wu H, Tao Y, Zhang B, Wang M, Mao F, Yan Y, Gao S, Gu H, Zhu W, Qian H. Exosomes released by human umbilical cord mesenchymal stem cells protect against cisplatin-induced renal oxidative stress and apoptosis in vivo and in vitro. Stem Cell Res Ther 2013; 4: 34 [PMID: 23618405 DOI: 10.1186/scrt194]

Tateishi-Yuyama E, Matsuura H, Uemura H, Ikeda U, Shintani S, Masaki H, Amano K, Kishimoto Y, Yoshimoto K, Akashi H, Shimada K, Iwasaka T, Imaizumi T. Therapeutic angiogenesis for patients with limb ischaemia by autologous transplantation of bone-marrow cells: a pilot study and a randomised controlled trial. Lancet 2002; 360: 427-435 [PMID: 12241713 DOI: 10.1016/S0140-6736(02)09070-8]

Abdel-Latif A, Bolli R, Tleyjeh IM, Montori VM, Perin EC, Hornung CA, Zuba-Surma EK, Al-Mallah M, Dawn B. Adult bone marrow-derived cells for cardiac repair: a systematic review and meta-analysis. Arch Intern Med 2007; 167: 989-997 [PMID: 17533201 DOI: 10.1001/archinte.167.10.9899]

Assmus B, Rolf A, Erbs S, Elässer A, Haberbosch W, Hambrecht T, Rillmanns H, Yu J, Corti R, Maltey DG, Harnn CW, Sülbecke T, Tomn T, Dimmeler S, Dill T, Zeiher AM, Schächinger V. Clinical outcome 2 years after intracoronary administration of bone marrow-derived progenitor cells in acute myocardial infarction. Circ Heart Fail 2010; 3: 89-96 [PMID: 19996415 DOI: 10.1161/CIRCHEARTFAILURE.108.843243]

Leistner DM, Fischer-Rasokat U, Honold J, Seeger FH, Schächinger V, Lehmann R, Martin H, Burck I, Urbanic C, Dimmeler S, Zeiher AM, Assmus B. Transplantation of progenitor cells and regeneration enhancement in acute myocardial infarction (TOPCARE-AMI): final 5-year results suggest long-term safety and efficacy. Clin Res Cardiovasc 2011; 100: 925-934 [PMID: 21639321 DOI: 10.1007/s00392-011-0327-y]

Walter DH, Krankenberg H, Balzer JO, Kalka C, Baumgartner I, Schlüter M, Tomn T, Seeger F, Dimmeler S, Lindhoff-Last E, Zeiher AM. Intravenous administration of bone marrow mononuclear cells in patients with critical limb ischaemia: a randomized-start, placebo-controlled pilot trial (PROVASC). Circ Cardiovasc Interv 2011; 4: 26-37 [PMID: 21205939 DOI: 10.1161/IJ]
Jeong JO, Han JW, Kim JM, Cho HJ, Park C, Lee N, Kim DW, Yoon YS. Malignant tumor formation after transplantation of short-term cultured bone marrow mesenchymal stem cells in experimental myocardial infarction and diabetic neuropathy. *Circ Res* 2011; **108**: 1340-1347 [PMID: 21493893 DOI: 10.1161/CIRCRESAHA.110.239848]

Noh H, Yu MR, Kim HJ, Jeon JS, Kwon SH, Jin SY, Lee J, Jang J, Park JO, Ziyadeh F, Han DC, Lee HB. Uremia induces functional incompetence of bone marrow-derived stromal cells. *Nephrol Dial Transplant* 2012; **27**: 218-225 [PMID: 21622994 DOI: 10.1093/ndt/gfr267]

Idziak M, Pędzisz P, Burdzińska A, Gala K, Pączek L. Uremic toxins impair human bone marrow-derived mesenchymal stem cells functionality in vitro. *Exp Toxicol Pathol* 2014; **66**: 187-194 [PMID: 24548687 DOI: 10.1016/j.etp.2014.01.003]

Klinkhammer BM, Kramann R, Mallau M, Makowska A, van Roeyen CR, Rong S, Buecher EB, Boor P, Kovacova K, Zok S, Denecke B, Stuettgen E, Otten S, Floege J, Kunter U. Mesenchymal stem cells from rats with chronic kidney disease exhibit premature senescence and loss of regenerative potential. *PLoS One* 2014; **9**: e92115 [PMID: 24667162 DOI: 10.1371/journal.pone.0092115]
