


Preservation Solutions for Kidney Transplantation: History, Advances and Mechanisms

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Abstract

Solid organ transplantation was one of the greatest medical advances during the past few decades. Organ preservation solutions have been applied to diminish ischemic/hypoxic injury during cold storage and improve graft survival. In this article, we provide a general review of the history and advances of preservation solutions for kidney transplantation. Key components of commonly used solutions are listed, and effective supplementations for current available preservation solutions are discussed. At cellular and molecular levels, further insights were provided into the pathophysiological mechanisms of effective ingredients against ischemic/hypoxic renal injury during cold storage. We pay special attention to the cellular and molecular events during transplantation, including ATP depletion, acidosis, mitochondrial dysfunction, oxidative stress, inflammation, and other intracellular mechanisms.

Keywords

kidney transplantation, preservation solutions, additives, ischemia, hypoxia, mechanism

Introduction

The 20th century had witnessed great advances in organ transplantation since the first human kidney transplant was attempted by Yuriy Voroniy in 1933¹. Kidney transplantation offers the opportunity to replace failed kidneys that are unable to perform their normal physiological functions. It is vital to preserve kidneys in optimal condition before transplantation takes place. The majority of kidney donations are now from non-heart-beating donors (NHBD), and it often takes hours, or even days under some circumstances, to transport organs between medical centers, with high risk of ischemic/hypoxic damage^{2,3}.

Numerous preservation solutions have been developed for organ storage to prevent ischemic/hypoxic injury, improve histocompatibility matches, and ensure immediate and normal organ function after implantation. Previous studies and clinical applications have demonstrated that the choices of different preservation solutions may affect both short-term and long-term transplant outcomes^{4,5}. To minimize ischemic damage, preservation solutions have been designed to specifically target against the biological changes that may occur during the ischemic/hypoxic process⁶. Pharmacological, biochemical, hormonal, and immunological approaches have been

applied in the development of preservation solutions (see previous reviews^{7–10}).

Based on our research experience, we will discuss in this review the history of several “gold-standard” preservation solutions, recent advances, and future developments in kidney preservation solutions. We highlight the underlying molecular mechanisms of commonly used preservation solutions against ischemic/hypoxic injury at present. Literature searches were conducted of online databases, including PubMed and MEDLINE, using kidney transplantation,

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preservation solutions, additives, ischemia, and hypoxia as keywords to capture all relevant publications. The final date for literature searches was July 15, 2019.

History

The importance of ensuring the preservation of kidneys in optimal condition between retrieval and implantation has been long recognized. Cold storage, in which a preservation solution is infused into the organ and the organ is stored at hypothermic conditions, is a process used to reduce the metabolic requirements of organs and attenuate ischemic/hypoxic injury. It has been successfully applied in kidney transplantation for more than 50 years, and for decades has been the most widely used organ preservation method due to its simplicity and low cost^{9,11}. Cold storage preservation solutions have continued to evolve since their early development. In the late 19th century, the concept of continuous perfusion and simple cooling with cold solutions was developed for organ preservation¹¹. In the 1930s, it was believed that continuous hypothermic perfusion could help eliminate toxic metabolic products and provide nutrients to preserved organs¹². In 1968, Belzer et al. performed a successful kidney perfusion preservation¹³. A year later, Geoffrey Collins developed the first cold storage preservation solution, named Collins Solution, which increased kidney preservation time up to 24 h¹⁴. Collins Solution was further modified by the Eurotransplant Foundation in 1980 into the Euro-Collins Solution (EC)¹⁵, which was commonly used in Europe for about 15 years. In the late 1970s, Ross et al. developed hyperosmolar citrate (HOC) solution (also known as Marshall's solution), which is effective for 72 h of kidney preservation and is still in clinical use today^{16,17}. In 1986, Belzer et al. reported a newly developed University of Wisconsin (UW) solution containing lactobionate and raffinose¹⁸. Another popular preservation solution in the 1980s was histidine-tryptophan-ketoglutarate (HTK) solution¹⁹. UW and HTK solutions were regarded as the "gold-standard" and accounted for the majority of preservation solutions used in the clinic^{9,20}. Celsior is an extracellular type preservation solution for cold storage preservation developed in 1994, and has also been widely used in kidney transplantation²¹.

Here, we concentrate on the most widely used solutions (EC, UW, HTK, Celsior, HC-A and IGL-1) with comparable post-transplant outcomes, and discuss several recent advances for kidney preservation solutions.

Commonly used solutions for kidney preservation

Collins Solution

Collins Solution preserved kidneys successfully with simple ice storage before transplantation^{14,22}. It was realized that kidneys can absorb water and sodium and lose potassium

when deprived of oxygen or nutrients under hypothermic conditions. Therefore, Collins Solution was designed to have high potassium, high magnesium, and low sodium concentrations to mimic the composition of intracellular fluid. It is a phosphate-based solution with high glucose concentration that balances the osmolarity¹⁴. Later, several modifications were made based on the Collins Solution, which became known as EC Solution¹⁵ (Table 1). EC was commonly used clinically for the next 15 years, until the "gold-standard" UW solution was introduced.

University of Wisconsin Solution

UW solution is the most common cold storage solution in organ preservation. It was initially developed as a preservation solution for pancreas transplantation¹⁸, but has been widely used in preserving different kinds of organs, including the kidney, liver, and small bowel, since the late 1980s²⁴⁻²⁶.

Like EC solution, UW is an intracellular preservation solution with low sodium and high potassium concentrations to mimic an intracellular environment¹⁸ (Table 1). There are several advantages of UW compared with EC: a) hydroxyethyl starch (HES) serves as the colloid carrier to minimize interstitial edema during perfusion; b) metabolic substrates such as lactobionate and raffinose were introduced to replace glucose to prevent cell swelling; c) allopurinol and glutathione were added as oxygen radical scavengers to reduce oxidative stress²⁷; d) adenosine is used as an adenosine triphosphate (ATP) precursor²⁸.

UW solution has proven superior to EC solution in both animal experiments and clinical trials²⁹⁻³¹. For example, in the dog kidney autotransplant model, all kidneys were viable when stored for 72 h in UW solution, while none were viable when stored for 72 h in EC solution, suggesting that UW solution effectively preserves kidneys for 72 h²⁹. The European Multicenter Trial in the 1990s demonstrated better kidney preservation outcomes using UW solution compared with EC solution³⁰, with fewer incidences of delayed graft function (DGF, 35.8% vs. 52.2%) and better 1-year graft survival (92% vs. 86%)³¹.

Despite its popularity, recent studies have suggested potential disadvantages of UW solution, including a) high viscosity generated by HES, which complicates tissue wash-out and saturation with preservation solution³²; b) addition of penicillin G, dexamethasone, and insulin before use²⁰; and c) the current cost per liter of UW solution is \$282, and the total cost of UW solution per liter is over \$300 when additives are included²⁰. Therefore, further modifications have been made to create more effective or economical solutions, such as RPS-96 (UW lacking HES)³³, dextran 40-based UW (replacing HES with dextran 40 in UW)³⁴, perfluorocarbons (PFC)-based UW³⁵, hyperbranched polyglycerol (HPG) solution (replacing HES with HPG in UW)³⁶, and sodium lactobionate sucrose (SLS) solution (replacing raffinose with sucrose in UW)³⁷.

Table 1. Composition of Common Kidney Preservation Solutions.

		EC ¹⁵	UW ¹⁸	HTK ¹⁹	Celsior ²¹	IGL-I ²³	HC-A ⁴⁹
Colloids (g/L)	Hydroxyethyl starch	—	50	—	—	—	—
	PEG-35	—	—	—	—	1	—
Impermeants (mM)	Glucose	198	—	—	—	—	—
	Lactobionate	—	100	—	80	100	—
	Mannitol	—	—	30	60	—	166
	Raffinose	—	30	—	—	30	—
Electrolytes (mM)	Na ⁺	10	30	15	100	120	80
	K ⁺	115	125	9	15	30	80
	Mg ²⁺	—	5	4	13	5	41
	Ca ²⁺	—	—	0.0015	0.26	0.5	—
	Cl ⁻	15	—	50	41.5	20	—
	SO ₄ ²⁻	—	5	—	—	5	41
	PO ₄ ³⁻	100	25	—	—	25	—
	HCO ₃ ⁻	10	—	—	—	—	—
	Citrate	—	—	—	—	—	55
	Anti-oxidants (mM)	Allopurinol	—	1	—	—	1
Glutathione		—	3	—	3	3	—
Nutrients (mM)	Tryptophan	—	—	2	—	—	—
	Adenosine	—	5	—	—	5	—
	Adenine	—	—	—	—	—	0.38
	Glutamic acid	—	—	—	20	—	—
	Histidine	—	—	198	30	—	—
	Ketoglutarate	—	—	1	—	—	—
Additives	Insulin (U/L)	—	40	—	—	—	—
	Penicillin G (U/L)	—	200,000	—	—	—	—
	Dexamethasone (mg/L)	—	16	—	—	—	—
Osmolality (mOsm/L)		406	320	310	360	320	380
pH		7.0	7.4	7.2	7.3	7.4	7.0
Cost (US\$/Liter)		275	282	181	228	184	153

Histidin-Tryptophan-Ketoglutarat Solution

HTK solution was originally developed and introduced by Bretschneider as a cardioplegia solution for open heart surgery in the 1970s¹⁹. Subsequently, it proved to be effective in preserving liver, kidney, and pancreas^{38,39}. Unlike UW solution, HTK solution is an extracellular preservation solution with high sodium and low potassium concentrations. Histidine, tryptophan, and ketoglutarate are the three key components in HTK solution. Histidine serves as a buffer and retards the tissue pH decline during cold ischemic/hypoxic conditions. Tryptophan acts as a free radical scavenger and membrane stabilizer. Another amino acid, ketoglutarate, serves as an energy substrate¹⁹ (Table 1). HTK solution has a viscosity similar to that of water. This lower viscosity provides a shorter cooling time to reach hypothermic conditions compared with UW solution, and also eliminates the need for pressurized perfusion.

The effectiveness of HTK solution was confirmed in clinical trials in the early 1990s. Eurotransplant conducted a randomized multicenter study comparing kidney graft preservation with HTK, UW, and EC solutions⁴⁰. The results showed the equivalence of HTK compared with UW in preservative ability, with an equal incidence of initial non-function and similar 3-year kidney graft survival^{24,41}.

A minimal DGF rate was also observed in HTK as compare with EC solution (0 vs. 33.3%)⁴².

However, Stewart et al. reported that if there are additional graft factors such as donation after cardiac death, HTK preservation was associated with an increased risk of graft loss with cold ischemia time over 8 h⁴³. Another potential disadvantage of HTK solution is the higher perfusion volume caused by its low viscosity. However, even with the greater volume of HTK needed, the overall cost is decreased compared with UW solution²⁰ (\$181/L vs. \$282/L, Table 1). Despite single critical papers, HTK solution is still one of the most popular kidney preservation solutions in use today.

Celsior Solution

Celsior is an extracellular type of preservation solution developed by Menasché et al. in 1994 for cold storage preservation of cardiac grafts²¹. Currently, it has been applied successfully to heart, lung, liver, pancreas, kidney, and small bowel preservation^{44,45}. Celsior is a high-sodium, low-potassium solution. It adopts the key effective components of both UW and HTK solution. Lactobionate and mannitol were used as impermeants to limit cellular edema. Reduced glutathione was added to Celsior solution as a free-radical scavenger. Celsior solution has greater buffering capacity

against acidosis and less viscosity than UW due to the absence of colloids (Table 1).

A multicenter randomized study of 187 renal transplants showed that the kidney preservation by Celsior solution was equivalent to that of UW solution in DGF rate (31.3% vs. 33.9%) and 2-year graft survival (84% vs. 75%)⁴⁶. The incidence of acute tubular necrosis was much lower in the Celsior group than in the UW group (23% vs. 36%) after transplantation⁴⁷. These results indicated equivalence, and even some advantages, of Celsior solution when compared with the traditional UW solution.

Hypertonic Citrate Adenine Solution

Hypertonic citrate adenine (HC-A) is the most widely used preservation solution for kidney transplantation in China. It was first developed in the late 1970s by Shanghai Changzheng Hospital and the Shanghai Blood Center⁴⁸. Since then, HC-A solution has been extensively applied in China for over three decades. HC-A is a hyperosmolar citrate-based solution with additive adenine as nutrient to improve kidney function after circulation (Table 1). HC-A solution has a high mannitol concentration to act as an impermeant to limit cellular edema and oxidative stress.

Twenty years after the application of HC-A solution, HC-A II solution was developed (based on the formula of HC-A) by Shanghai Changzheng Hospital. HC-A II has a double-buffer system of citrate and phosphate to achieve enhanced buffer capacity. The concentration of magnesium was significantly reduced, and concentration of adenosine was increased compared with the HC-A solution. New ingredients of arginine, tryptophan, and ligustrazine were also added to stabilize cell membranes and protect mitochondrial function⁴⁹. In a multi-center randomized controlled trial conducted from 2008 to 2012 in China, HC-A II solution showed similar efficacy and safety in kidney preservation compared with HTK solution, suggesting that HC-A II solution is a promising option for clinical application⁴⁹.

Application of HC-A solution accounts for approximately 50% of total kidney transplantation in China nowadays due to its efficacy and low cost, although few specific studies on HC-A have been reported in the English literature. Indeed, the efficacy and safety of HC-A solution seem reasonably sound since it has been used for more than 100,000 grafts in clinical settings in China⁵⁰. However, more studies are needed due to lack of detailed data from basic and clinical research.

Other Solutions

Apart from the above mentioned solutions, several other solutions are also utilized for cold storage preservation of kidneys, including phosphate buffered sucrose (PBS) 140^{51,52}, HP16⁵³, HBS⁵⁴, B2⁵⁵, Lifer^{56,57}, Ecosol⁵⁸, Biolasol^{59,60}, renal preservation solution 2 (RPS-2)⁶¹, F-M⁶², AQIXRS-I⁶³, WMO-II⁶⁴, CZ-1⁶⁵ solutions, etc. Each

solution may vary in composition; however, the key strategy with each solution is similar: use colloids, impermeants, electrolytes, antioxidants, and nutrients to minimize ischemic/hypoxic injury and maximize kidney function after reperfusion^{66,67}.

Recent Advances in Kidney Preservation Solutions

Due to a deeper understanding of the process of ischemic/hypoxic injury and components of commonly used preservation solutions, efforts have been made to develop new preservation solutions. Particular components are now added to standard solutions to further prevent organs from ischemic/hypoxic injury. In the present review, we attempt to provide a summary of the effective additives available, with experimental evidence of animal models (Table 2).

Colloids

Several high molecular weight colloids have been applied in preservation solutions to sustain the intravascular oncotic pressure and prevent interstitial edema¹³¹. UW solution uses HES (50 kDa), which is a stable non-toxic oncotic agent, as the colloids¹³². Moreover, other colloids were also utilized to substitute HES to make solutions more cost-effective.

Dextran (40 kDa) is one of the most widely investigated replacements of HES³⁴. It is a safe replacement of HES in UW solution for the purpose of clinical kidney preservation³⁴. Dextran 40 may be beneficial during the graft reperfusion phase due to reduced vascular resistance and anti-thrombogenesis function^{34,133}.

Poly-ethylene glycol (PEG), a neutral water soluble non-toxic polymer, has been used to substitute HES in the several newly developed preservation solutions¹³⁴ including Institute Georges Lopez-1 (IGL-1), Polysol, and "Solution de Conservation des Organes et des Tissus" (SCOT) solution. IGL-1 is a PEG 35 kD-based preservation solution with inverted Na⁺/K⁺ concentration compared with that of UW solution¹³⁵ (Table 1). Encouraging results of IGL in kidney transplantation have been demonstrated in both rat and pig transplantation studies^{68,136}. In a multi-center study conducted in 2009, IGL-1 solution-preserved kidneys showed outcomes similar to those preserved in the UW solution in terms of DGF, rejection rates, and patient and graft survival¹³⁷. Polysol is another PEG 35 kD-based preservation solution that contains antioxidants, amino acids, energy substrates, and vitamins¹³⁸. In pig kidney transplant studies, Polysol displayed improved microcirculation and structural integrity following prolonged ischemia compared with UW and HTK solutions¹³⁹⁻¹⁴¹. Nonetheless, a pilot clinical study failed to demonstrate the advantage of Polysol in living kidney transplantation, with higher acute rejection rates observed in recipients compared with UW solution controls¹⁴². Therefore, the use of Polysol solution in the clinic is questionable. SCOT is another PEG-based preservation

Table 2. Recent Advances in Kidney Preservation Solutions.

Effective additives	Solutions	Species	Target pathway(s)	References
PEG	IGL-I, polysol, SCOT, PBSc+PEG, SBS+PEG, EC+PEG, UW+PEG	rat, pig, rabbit	intravascular oncotic pressure	68,69–74
dextran	HTK-N+dextran, dextran-based UW	pig, human	intravascular oncotic pressure	34,75
H ₂ S	UW+H ₂ S	rat	necrosis, apoptosis, inflammation, mitochondrial function	76,77
H ₂	UW+H ₂	rat	oxidative stress, apoptosis, fibrosis	78
CO	UW+CO, UW+CORM-3	rat, pig	oxidative stress, vasoconstriction, inflammation, apoptosis	79–81,82
O ₂ carrier	UW+M101, HTK+M101	pig	inflammation, fibrosis	83
Ar	CS+Ar	pig	inflammation, necrosis, fibrosis, apoptosis	84
endothelial receptor antagonist	UW+ET _A /ET _B receptor antagonists, EC+ET _A /ET _B receptor antagonists	rat	vasoconstriction	85–88
rho-kinase inhibitor HA1077	HTK+HA1077	pig	vasoconstriction	89
melagatran	UW+melagatran	pig	inflammation, fibrosis	90–92
trophic factors	UW+[BNP-I, SP, NGF-β, IGF-I, EGF], EC+HGF, EC+IGF-I	pig, dog	inflammation, fibrosis, apoptosis	93–95,96,97
rh BMP-7	saline+rh BMP-7	rat	oxidative stress	98
TNF-RFP	HTK+ TNF-RFP	pig	inflammation	99
cyclic helix B peptide	HOC+cyclic helix B peptide	pig	apoptosis, inflammation	100
L-arginine	HTK+L-arginine	dog	oxidative stress	101
caspase-3 siRNA	HOC+caspase-3 siRNA	pig	apoptosis, acidosis	102
MMP-2 siRNA	KPS-I+MMP-2 siRNA	rat	fibrosis	103
ICAM-1 antisense oligodeoxynucleotide IP-9125	EC+IP-9125	rat	inflammation	104
taurine	UW+taurine	pig	Ca ²⁺ homeostasis	105,106
ranolazine	PBSI40+ranolazine	pig	Ca ²⁺ homeostasis	107
verapamil	EC+verapamil	rat	Ca ²⁺ homeostasis	108–110
TMZ	EC+TMZ, UW+TMZ	pig	inflammation, mitochondrial function, oxidative stress	111–116
trolox, deferoxamine, ascorbate	UW+trolox, UW+deferoxamine, UW+ascorbate	rabbit	oxidative stress	117
lec-SOD	HOC+lec-SOD	rat	oxidative stress, apoptosis, inflammation	118
edaravone	HTK+edaravone	dog	oxidative stress	119
MitoQ	UW+MitoQ	pig, rat	mitochondrial function, oxidative stress	120,121
quinacrine	UW+quinacrine	rabbit	mitochondrial function	122
N-acetylcysteine, sodium nitroprusside, phosphoramidon	N-acetylcysteine+sodium nitroprusside+phosphoramidon	rat	oxidative stress	123
selenium	HTK+selenium	pig	oxidative stress	124,125
nicaraven	UW+ nicaraven, EC+ nicaraven	rat, dog	oxidative stress	126
propofol	HTK+propofol	pig	oxidative stress	127
prostaglandin E1	UW+prostaglandin E1	human	oxidative stress	128,129
tanshinone IIA	CS+tanshinone IIA	rat	oxidative stress, apoptosis	130

PEG, polyethylene glycol; CORM, carbon monoxide releasing molecules; BNP-I, bovine neutrophil peptide-I; SP, substance P; NGF, nerve growth factor; IGF-I, insulin-like growth factor-I; EGF, epidermal growth factor; HGF, hepatocyte growth factor; rh BMP-7, recombinant human bone morphogenetic protein-7; TNF-RFP, TNF-receptor fusion protein; MMP, matrix metalloproteinase; ICAM, intercellular adhesion molecule; TMZ, trimetazidine; lec-SOD, lecithinized superoxide dismutase; MitoQ, mitoquinone; IGL-I, Institute Georges Lopez-I solution; SCOT, Solution de Conservation des Organes et des Tissus; PBSc, Phosphate-buffered sucrose solution; SBS, sucrose-based solution; EC, Euro-Collins solution; UW, University of Wisconsin solution; HTK, Histidin-Tryptophan-Ketoglutarat solution; CS, Celsior solution; HOC, hyperosmolar citrate solution; KPS-I, kidney perfusion solution-I.

solution, which contains PEG 20kD as a colloid¹⁴³. The effectiveness of SCOT in kidney transplantation was displayed in a preliminary clinical study, with comparable or better outcomes versus commonly used UW or EC solutions¹⁴⁴.

PEG could spontaneously bind to cell surfaces to stabilize membrane lipids^{145,146} and may further enhance immunoprotective properties^{134,147}. It has vasodilatory, antifibrosis, and antioxidant effects in addition to the colloid

properties^{134,135}. Thus, PEG-based preservation solutions are considered promising alternatives for commonly used solutions in clinical applications.

Gases

The use of therapeutic gases as effective additives in the context of preservation solutions has become popular in recent years¹⁴⁸. Several gases have been used to reduce ischemic/hypoxic injury in kidney transplantation, including oxygen (O₂), hydrogen (H₂), carbon monoxide (CO), nitric oxide (NO), hydrogen sulfide (H₂S) and argon (Ar).

The retrograde venous application of humidified pure O₂ during cold storage was developed by Isselhard et al. in 1972¹⁴⁹. The effect was further confirmed by a small pilot clinical study¹⁵⁰. In a large animal preclinical model, addition of M101 (an O₂ carrier with high-oxygen affinity) into UW and HTK solutions provided significant benefits to grafts, both in early function recovery and long-term outcomes⁸³.

H₂ is a highly diffusible antioxidant gas, which can easily enter the cell. H₂ could preserve mitochondrial function by attenuating reactive oxygen species (ROS) production, thus preventing apoptotic cell death¹⁵¹. A rat study has demonstrated the beneficial effect of H₂-rich UW solution during kidney transplantation. H₂-rich UW solution showed decreased oxidative stress, tubular apoptosis, interstitial fibrosis, improved renal function and survival rate compared with UW simple cold storage⁷⁸.

CO, NO, and H₂S are considered to be endogenous gas transmitters. CO has been implicated to have cytoprotective effects by attenuating proinflammatory cytokines, lipid peroxidation, and improving mitochondrial function^{152,153}. Supplementation of UW solution with CO or CO-releasing molecules provided significant protection against renal ischemic/hypoxic injury in both rat and porcine transplantation studies^{79–81}. NO is mainly generated from the endogenous metabolism of L-arginine to citrulline by NO synthase¹⁵⁴. The therapeutic application of NO has been implicated in attenuating ischemia-reperfusion injury¹⁵⁵. Therapeutic effects of H₂S have also been demonstrated in animal models of ischemia and reperfusion^{156,157}. H₂S treatment improved early allograft function and survival by attenuating the necrosis and apoptosis of glomerular and tubular cells^{76,77}.

In addition, argon saturated Celsior solution (Ar-Celsior) decreased inflammation, interstitial fibrosis, tubular necrosis, and apoptosis in a porcine kidney transplantation model, resulting in improved early functional recovery, graft quality, and survival⁸⁴. Therefore, adding gas at an appropriate level and composition into current preservation solutions is a promising approach to improve kidney preservation.

Amino Acids, Peptides and Proteins

Amino acids appear to be quite effective in providing nutrients or acting as metabolic substrates. The commonly used

preservation solutions utilize amino acids, such as tryptophan in HTK solution, glutamic acid in Celsior solution, and histidine in both HTK and Celsior solution, as energy substrates (Table 1). Amino acids with antioxidant effects were also introduced into newly developed preservation solutions as additives. L-arginine is the endogenous substrate for NO synthase. In an animal model of canine kidney transplantation, L-arginine-enriched preservation solution presented a beneficial and protective effect on long-term (72 h) hypothermic ischemic damage, with reduced lipid peroxidation¹⁰¹. In a rat model, pre-administration of N-acetylcysteine attenuates renal ischemia-reperfusion injury by protecting cells against free radical damage¹²³. Oral administration of D-cysteine attenuates renal ischemia-reperfusion injury by producing H₂S¹⁵⁸. Addition of Cyclic Helix B peptide into preservation solutions ameliorated renal injury after porcine kidney transplantation, which may be associated with decreased apoptosis and inflammation¹⁰⁰.

Recent studies have focused on trophic factors as additives to improve storage efficiency of preservation solutions^{93,94,159}. With the addition of a combination of bovine neutrophil peptide-1 (BNP-1), substance P (SP), nerve growth factor-β (NGF-β), insulin-like growth factor-1 (IGF-1), and epidermal-like growth factor (EGF) to the UW solution, McAnulty et al. successfully achieved 6-day kidney preservation in a canine model⁹³. Moreover, addition of hepatocyte growth factor (HGF) to EC solution accelerated both renal blood flow recovery and glomerular filtration rate in dog kidney during cold ischemia⁹⁵. Experimental evidence has suggested that trophic factors protect cells against inflammation, fibrosis and apoptosis, and improve mitochondrial function as well¹⁶⁰.

Recombinant human bone morphogenetic protein-7 (rh BMP-7) perfused rat kidneys had elevated superoxide dismutase (SOD) activity and decreased lipid peroxidation level versus the UW-treated group⁹⁸. The same effect was also achieved by direct introduction of lecithinized superoxide dismutase (lec-SOD) into the preservation solution¹¹⁸. TNF-receptor fusion protein (TNF-RFP) was able to suppress inflammation and promote organ survival in xenoperfused porcine kidneys⁹⁹.

Signaling Pathway Blockers

The use of siRNAs, oligonucleotides, specific inhibitors or antagonists represented effective approaches to specifically target signaling pathways involved in ischemic/hypoxic injury. Exogenous administration of naked siRNAs via preservation solutions have been successfully tested in animal studies. Introducing naked caspase-3 siRNA in preservation solution resulted in reduced caspase-3 expression levels, cell apoptosis, and better acid-base homeostasis in a porcine kidney transplantation model¹⁰². Similarly, supplementation of preservation solution with matrix metalloproteinase (MMP)-2 siRNA protected transplant rat kidneys from

preservation injury by inhibiting MMP-2 expression and tissue fibrosis¹⁰³.

The adhesion of immune competent cells to the endothelial wall represents an important aspect of ischemic/hypoxic injury. Thus, phosphorothioate intercellular adhesion molecule (ICAM)-1 antisense oligodeoxynucleotide was added to EC solution to reduce ICAM-1-related inflammation response¹⁰⁴. There was also experimental evidence of the benefit of blocking the vasoconstrictor mediators in improving kidney function. Studies in rats have shown that endothelial receptor antagonists could ameliorate renal ischemic/hypoxic injury after transplantation^{85–88}. Supplementation of Rho-kinase inhibitor HA1077 could improve renal function by antagonizing vasoconstriction⁸⁹. Supplementing UW solution with a thrombin inhibitor melagatran in a porcine kidney transplantation model resulted in strong protection against both acute renal injury⁹⁰ and chronic lesions, including interstitial fibrosis, apoptosis, and inflammation^{91,92}. Calcium channel blocker verapamil^{108–110} and platelet-activating factor (PAF) receptor antagonist¹⁶¹ were also candidate additives in preservation solutions, the effect of which were confirmed in animal models. The postulated benefits include reduction of calcium overload, increase in tissue ATP concentration, and sodium re-absorption. Application of proteasome inhibitor is a potential strategy to reduce ischemic/hypoxic injury in both liver and heart transplantation, which lead to the up-regulation of AMPK activity, down-regulation of mTOR signaling, and influence autophagy^{162,163}. Thus, supplying proteasome inhibitors into common preservation solutions can become a strategy to decrease renal ischemic/hypoxic injury as well. In conclusion, pathway-specific blockers that are based on well-defined ischemic/hypoxic mechanisms offer good opportunities to improve commonly used preservation solutions.

Other Pharmacological Additives

Apart from above-mentioned supplements, other pharmacological additives including radical scavengers, mitochondrial protectors and drugs have also been added to preservation solutions. The addition of radical scavengers (nicaraven¹²⁶, trolox¹¹⁷, edaravone¹¹⁹) could trap free radicals and protect cells against oxidative stress directly, contributing to reduced lipid peroxidation and higher survival rate of the preserved kidneys. In addition, exogenous administration of selenium^{124,125}, propofol¹²⁷ (a widely used anesthetic), and tanshinone IIA¹³⁰ (an effective component of the traditional Chinese medicine Danshen) in preservation solutions tended to have antioxidant effects to attenuate malondialdehyde concentration, followed by the improvement in long-term renal function after transplantation.

Mitoquinone (MitoQ)^{120,121} and quinacrine¹²² act as mitochondrial targeted antioxidant compounds to reduce the damage of mitochondrial electron transport chain and improve renal cell viability in rat and porcine studies.

Trimetazidine (TMZ) is another drug with mitochondrial-protective¹⁶⁴ functions. Supplementation of TMZ in UW solution was beneficial in porcine models, with improved functional recovery of preserved kidneys, higher survival rate, as well as reduced cellular and mitochondrial swelling, immune cell invasion, and interstitial fibrosis^{111–116}.

Other pharmacological additives that were beneficial in animal studies including prostaglandin E1^{128,129}, taurine^{105,106}, ranolazine¹⁰⁷, etc. (Table 2). They also have the potential to be used in clinical transplantation. However, the underlying mechanisms and the specific signaling pathways involved are not well understood yet. Thus, more experimental studies and further clinical trials are still needed.

Mechanisms of Preservation Solutions Against Ischemic/Hypoxic Renal Injury

Ischemia and hypoxia are inevitable events during cold preservation of kidney transplantation. Ischemic/hypoxic renal injury is caused by a sudden cessation of blood flow along with the immediate oxygen deprivation to the kidneys¹⁶⁵. Tissues are starved of oxygen and nutrients, leading to the accumulation of metabolic waste products. The imbalance in metabolic supply and demand within the ischemic organ results in profound tissue hypoxia and dysfunction. Subsequent reperfusion further enhances the activation of immune responses and cell death programs. A wide range of pathophysiological processes have been identified to be related to ischemic/hypoxic renal injury during cold storage, including ATP depletion, calcium overload, acidosis, mitochondrial dysfunction, oxidative stress, inflammatory response, and eventual cell death programs^{166,167} (Fig. 1).

ATP Depletion and Loss of Electrolytes Homeostasis

Nearly 95% of cellular ATP is depleted within 4 h as a consequence of organ cold storage during transplantation⁶. On the one hand, ATP deficits lead to a reduced activity of Na⁺/K⁺ ATPase, thereby contributing to increased intracellular sodium concentration¹¹. Large amounts of water enter the cytoplasm due to the hyperosmolar intracellular environment, resulting in cell edema¹⁶⁸. On the other hand, Na⁺/Ca²⁺ exchangers begin to work in the reverse direction and stop pumping calcium out of the cell, leading to calcium accumulation in cells¹⁶⁹. Increased intracellular calcium levels can activate calcium-dependent phospholipase and proteases, promoting cell apoptosis¹⁶⁶ (Fig. 1).

To combat the above-mentioned ischemic/hypoxic renal injury during cold storage, a number of approaches have been used in preservation solutions. Energy substrates such as adenosine were often provided in solutions to allow for rapid ATP regeneration during preservation. The calcium channel blocker verapamil^{108–110} and other pharmacological reagents, which can be beneficial to Ca²⁺ homeostasis^{105–107}, are added into preservation solutions

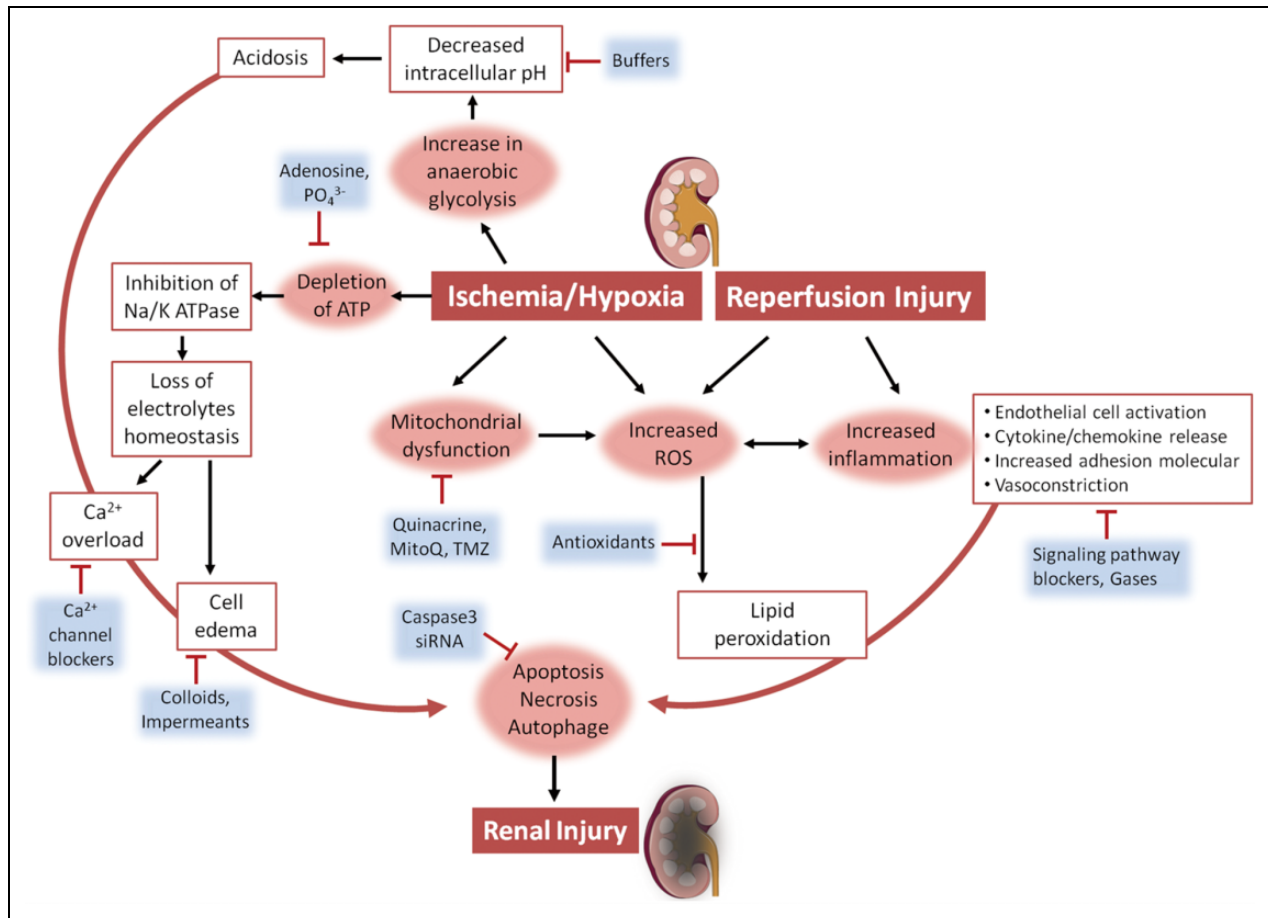


Fig 1. Schematic representation of effective supplementations in preservation solutions against ischemic/hypoxic renal injury.

to prevent calcium overload (Table 2). H_2S could inhibit Na^+/H^+ exchanger activity via the PI3K/Akt/PKG-dependent pathway, hence reducing Ca^{2+} overload¹⁷⁰. Impermeants and colloids are added to preservation solutions to prevent cell swelling¹⁷¹. The effectiveness of impermeants in preventing cell swelling is determined by their molecular weight (MW), with larger molecules being more effective. The monosaccharide glucose (MW 180) was applied in EC solution in the early stages but was soon abolished because it could pass through the cell membrane and become a source of lactate under hypoxic conditions¹⁰. The slightly larger monosaccharide mannitol (MW 182), which also has antioxidant potential, was used to replace glucose in HTK, Celsior, and HA-C solutions. Sucrose (MW 342) is a disaccharide and was used in phosphate-buffered sucrose (PBS) solution¹⁷². Raffinose (MW 504) was added to UW solution, along with lactobionate (Table 1). Colloids such as HES¹⁸, PEG^{134,68,136,142}, and dextran^{34,133} were also common components in preservation solutions, with convincing evidence of anti-edema function (see section *Colloids* for details).

Acidosis

Under ischemic/hypoxic conditions, energy metabolism in cells switches from fatty acid oxidation to more oxidation-efficient anaerobic glycolysis, allowing organs to sustain cellular viability¹⁷³. Decreased intracellular pH and acidosis are the immediate results of anaerobic glycolysis, and are often observed in ischemic tissues. Mild acidosis has been suggested to favor cell survival by inhibiting activated proteases and phospholipases via the rate-limiting step in glycolysis. However, strong activation of proteases and phospholipases by severe acidosis could lead to protein and lipid breakdown, lysosomal damage, and eventual cell death¹⁷⁴. Therefore, adequate control of cellular pH has become an important function of preservation solutions¹⁷⁵. To achieve this goal, different buffering systems were applied. EC and UW solutions used phosphate as a buffer, while HTK and Celsior solutions used histidine (Table 1). Among these commonly used preservation solutions, HTK has the highest buffering capacity due to a high concentration of histidine¹⁷⁵. The pH of common preservation solutions is between 7.0 and 7.4.

Mitochondrial Dysfunction

Mitochondrial dysfunction has been regarded as a critical event during ischemia. Under ischemic/hypoxic conditions, oxidative phosphorylation in the mitochondria is suppressed by a lack of oxygen, leading to impaired ATP synthesis and further severe ATP depletion¹⁷⁶. There is experimental evidence that mitochondrial Ca^{2+} uptake was dramatically increased during cold ischemia, especially when it is challenged by high extra-mitochondrial Ca^{2+} concentrations¹⁷⁷, resulting in impaired mitochondrial structure and function. Mitochondrial dysfunction may influence energy regeneration at the re-oxygenation stage¹⁷⁸. Mitochondria are the major source of intracellular ROS production¹⁷⁶. The uncoupling of the mitochondrial respiratory chain induces the formation of ROS, a process which is enhanced during the reperfusion stage. This could contribute to the denaturation of proteins, nucleic acids, and lipids, which induce cell apoptosis or necrosis¹⁷⁶.

It is thus clear that maintaining mitochondrial integrity and protecting mitochondria function are key principles in developing preservation solutions. The supplementation of preservation solutions with mitochondrial-protective reagents H_2S , MitoQ^{120,121}, quinacrine¹²², and TMZ¹¹¹⁻¹¹⁶ has proven to be effective (Table 2). AP39, a novel mitochondria-targeted H_2S donor, can also stimulate cellular bioenergetics, and protect against the loss of mitochondrial DNA integrity¹⁷⁹.

Oxidative Stress

The reperfusion stage, which occurs hours or days after the initial ischemic/hypoxic insult, is regarded as the final stage of ischemic injury¹⁶⁸. It has a profound influence not only on the short-term but also the long-term recovery outcome of a transplanted kidney^{168,180}. During this stage, blood flow and oxygen are re-introduced into organs, leading to a burst of ROS¹⁸¹. Hydrogen peroxide (H_2O_2) and the superoxide anion (O_2^-) are mainly generated by xanthine oxidase, and further lead to hydroxyl radical (OH^\cdot) formation. Meanwhile, cold storage itself has been suggested to promote ROS production via mitochondrial dysfunction^{182,183}. ROS react rapidly with other molecules, leading to lipid peroxidation and oxidative damage of nucleic acids and proteins¹⁸⁰, and eventually contribute to cell apoptosis¹⁷⁶ (Fig. 1).

Thus, inhibiting ROS production, especially at times of reperfusion, has become a key strategy to protect organs during transplantation. The beneficial effects of antioxidants and radical scavengers against ischemic/hypoxic injury have been confirmed by numerous studies. In UW solution, allopurinol (a xanthine oxidase inhibitor) and reduced glutathione (a thiol containing amino acid) were included to reduce ROS formation. Similarly, HTK solution contains tryptophan and histidine as ROS-scavenging amino acids (Table 1). Experimental additions of antioxidant agents into common preservation solutions have proven to be effective.

Direct introduction of lecithinized superoxide dismutase (lec-SOD), a catalyzer of ROS degradation, into preservation solution demonstrated a long-term benefit of reducing oxidant stress¹⁸⁴. H_2S has also been studied for its anti-oxidative properties. The benefits were possibly related to lowering the generation of free radicals, inducing antioxidant gene expression and anti-apoptotic functions^{82,185}. N-acetylcysteine contains a thiol group, which is readily accessible to the then intracellular compartment and could scavenge free radicals¹²³. As a widely used anesthetic, propofol displays its antioxidant activity through reduced lipid peroxidation and increased SOD levels¹²⁷. Moreover, several other compounds have also been added to preservation solutions, and were described to protect grafted kidneys against ischemic/hypoxic injury through direct or indirect antioxidative stress properties, including TMZ¹⁸⁶, rh-BMP-7⁹⁸, L-arginine¹⁰¹, trolox¹¹⁷, edaravone¹¹⁹, selenium¹²⁵, nicaraven¹²⁶, prostaglandin E1^{128,129}, and tanshinone IIA¹³⁰ (Table 2). Taken together, these findings support the idea that interrupting the ROS generation pathway and scavenging existing ROS could be effective strategies in preventing ischemic/hypoxic injury during kidney preservation (Fig. 1).

Inflammation Process

The inflammatory response is another profound consequence of blood flow restoration and re-oxygenation in the reperfusion stage¹⁷¹. ROS generation and lipid peroxidation process seem to be major factors in the activation of innate immunity^{166,187}. Inflammatory cells express NADPH oxidase, which in turn leads to the formation of additional ROS, further potentiating renal damage¹⁸⁸ (Fig. 1).

During reperfusion, activation of endothelial cells trigger the downstream signaling pathways, including mitogen-activated protein kinase (MAPK) and NF- κ B, leading to the release of pro-inflammatory cytokines (e.g. IL-6 and TNF- α) and chemokines^{166,189}. Chemokines are regarded as major mediators of inflammation that regulate adhesion molecules expression as well as leukocyte infiltration and activation¹⁷⁶. Increased expression of adhesion molecules and Class II MHC proteins result in vasoconstriction, exacerbating tissue damage. Increased platelet activation and adhesion to the injured vascular endothelium also reduces blood flow and exacerbates ischemia^{167,190}.

Multiple inflammatory pathways and factors take part in this process. Therefore, specific antibodies or pathway inhibitors have been applied in preservation solutions to modulate the inflammatory responses and attenuate post-ischemic injury. Direct inhibition of endothelial cell activation and vasoconstriction can be achieved by using endothelial receptor antagonists in preservation solutions⁸⁵⁻⁸⁸. The pro-inflammatory effects of TNF- α were effectively blocked by application of TNF-receptor fusion protein (TNF-RFP)⁹⁹. Administration of ICM-1 antisense deoxynucleotides ameliorated inflammatory injury⁹⁴. Supplementation of UW solution with FR167653, a specific p38MAPK inhibitor,

showed important benefits to graft function by reducing inflammation⁹⁴. Administration of exogenous CO or CO-releasing molecules has cytoprotective effects involving anti-inflammatory response and hypoxia-inducible factor (HIF)-1 α stabilization¹⁹¹. In addition, agents like H₂S^{156,76}, O₂⁸³, Ar⁸⁴, melagatran^{90,91}, cyclic helix B peptide¹⁰⁰, TMZ^{111–116}, and lec-SOD¹¹⁸ have also been implicated in preventing tissue inflammation during ischemia and reperfusion in various experimental models (Table 2 and Fig. 1).

Cell Apoptosis, Necrosis and Autophagy

Ischemic/hypoxic and subsequent reperfusion injury leads to the activation of cell death programs, which can be categorized as apoptosis, necrosis, and autophagy-associated cell death¹⁹². Apoptosis is a process of programmed cell death that is characterized by blebbing, cell shrinkage, loss of mitochondrial integrity, and nuclear fragmentation¹⁹³. Cell apoptosis may be induced by mitochondrial dysfunction. Cytochrome *c* was released into the cytosol from damaged mitochondria to induce the formation of apoptosomes. Subsequent apoptotic nuclear DNA damage was induced by the release of apoptosis-inducing factor (AIF) and endonuclease G^{168,194}. Ischemic/hypoxic mechanisms such as oxidative stress and inflammatory signaling may contribute to apoptosis as well. Thus, blocking these pathways to inhibit apoptosis may have promises as therapeutic strategies for ischemic/hypoxic renal injury during cold storage. In addition, apoptosis can be blocked by direct target apoptosis effectors. The glucocorticoid, dexamethasone, has an inhibitory effect against apoptosis by upregulating Bcl-XL, inhibiting Bax production, and activating caspases-3/9 in renal ischemia-reperfusion injury rodent models¹⁹⁵. Administration of naked caspase-3 siRNA in a preservation solution resulted in reduced caspase-3 expression level and cell apoptosis, elucidating the protective effect in isolated ischemic porcine kidneys¹⁰².

Necrosis, characterized by progressive cell swelling, plasma membrane rupture, and leakage of proteases and lysosomes into the extracellular compartment, is a frequent outcome of ischemic/hypoxic renal injury during cold storage¹⁹². Strong innate inflammatory signaling leads to cell necrosis, while necrosis in turn promotes inflammation through leakage of cellular contents, inflammatory cell infiltration, and cytokine production¹⁹⁶.

Unlike necrosis, autophagy is a regulated, programmed process that disassembles unnecessary or dysfunctional components of the cell¹⁹². Autophagy-associated cell death is also a common outcome after renal cells are exposed to ischemic/hypoxic conditions. However, whether renal cells undergo necrosis or programmed cell death appears to be ATP-concentration dependent. Studies have shown that ATP is required for programmed cell death, whereas complete ATP depletion would lead to necrosis^{186,197}. Experimentally, the effectiveness of pharmacological additives that may reduce cell swelling or ATP depletion on preventing

necrosis have been recently illustrated (Table 2). AMP-activated protein kinase (AMPK) is an energy sensor that regulates cellular metabolism and homeostasis. Renal ischemia was reported to induce AMPK phosphorylation, and sequentially promotes autophagy through phosphorylation of downstream effectors. Treatment of rat or mouse models with AMPK activator attenuates renal ischemia/reperfusion injury, with decreased tubular necrosis and expression of apoptosis-related proteins, improved glomerular filtration rate, and increased expression of autophagy-related proteins^{198,199}.

Conclusion and Perspectives

There has been major progression in developing preservation solutions for kidney transplantation in the past decades. To date, thousands of kidney transplants have been successfully performed using conventional preservation solutions. The main principle of organ preservation solution is to maintain the organ viability to the greatest extent during storage to achieve better transplant outcomes after the reperfusion stage²⁰⁰. The principal ingredients of preservation solutions, colloids, buffers, nutrients, and antioxidants are responsible for counteracting cellular edema, acidosis, ATP depletion, and the production of ROS, respectively. With a better understanding of the mechanisms of ischemic/hypoxic renal injury, it is now well recognized that supplementation of preservation solutions with pharmacological molecules has the great potential to improve graft quality. Pharmacological additives that specifically target ROS, inflammation, and apoptosis signaling pathways have proven to be effective.

UW, HTK, and Celsior solutions displayed a much lower risk of DGF after kidney transplantation compared with traditional EC solutions. The incidence of DGF is equal among these three solutions according to randomized clinical trials⁹. Thus, these well-established and commonly used solutions account for the majority of preservation solution used in the clinic. Meanwhile, numerous new preservation solutions with supplemental pharmacological agents have shown promising effects in ameliorating ischemic/hypoxic renal injury and DGF in animal studies, but clinical evidence is still lacking. Challenges remain in their translation from the bench to the bedside, including the specific dosage of additives, method of drug delivery, and potential off-target effects.

Since ischemic/hypoxic renal injury involves multiple mechanisms, targeting a single molecule or a single signaling pathway is unlikely to provide a complete amelioration of ischemic/hypoxic renal injury during cold storage. Instead, multi-drug approaches or a single drug targeting multiple molecules and/or pathways will be a more reliable strategy for better preservation of renal graft. The utilization of multi-drugs may activate protective signaling pathways and inhibit damaging ones at the same time. Indeed, several studies have explored the combination effects of compounds, each targeted against a specific pathway, and have

demonstrated additivity of the approaches^{94,123}. It still remains urgent to gain additional mechanistic insights into the molecular events and the underlying mechanisms related to ischemic/hypoxic renal injury during cold storage. The future development of new preservation solutions would be accelerated by the modern drug industry, with the systematic analyzing of dysregulated genes during ischemic/hypoxic damage and the screening of new drugs. Despite the challenges ahead, we are hopeful that new preservation solutions for kidney transplantation against ischemic/hypoxic renal injury will soon be applied for clinical practice through joint efforts of worldwide basic and clinical researchers, including our research team.

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References

- Matevossian E, Kern H, Huser N, Doll D, Snopok Y, Nahrig J, Altomonte J, Sinicina I, Friess H, Thorban S. Surgeon Yurii Voronoy (1895-1961) - a pioneer in the history of clinical transplantation: in memoriam at the 75th anniversary of the first human kidney transplantation. *Transpl Int*. 2009;22(12):1132–1139.
- Alvarez J, del Barrio R, Arias J, Ruiz F, Iglesias J, de Elias R, Yebenes C, Matesanz J, Caniego C, Elvira J. Non-heart-beating donors from the streets: an increasing donor pool source. *Transplantation*. 2000;70(2):314–317.
- Daemen JW, Oomen AP, Kelders WP, Kootstra G. The potential pool of non-heart-beating kidney donors. *Clin Transplant*. 1997;11(2):149–154.
- Opelz G, Dohler B. Multicenter analysis of kidney preservation. *Transplantation*. 2007;83(3):247–253.
- Stewart ZA, Lonze BE, Warren DS, Dagher NN, Singer AL, Montgomery RA, Segev DL. Histidine-tryptophan-ketoglutarate (HTK) is associated with reduced graft survival of deceased donor kidney transplants. *Am J Transplant*. 2009;9(5):1048–1054.
- Maathuis MH, Leuvenink HG, Ploeg RJ. Perspectives in organ preservation. *Transplantation*. 2007;83(10):1289–1298.
- Saint Yves T, Delpech PO, Giraud S, Thuillier R, Hauet T. Additives to preservation solutions. *Prog Urol*. 2014;24(suppl 1):S31–S36.
- Chatauret N, Thuillier R, Hauet T. Preservation strategies to reduce ischemic injury in kidney transplantation: pharmacological and genetic approaches. *Curr Opin Organ Transplant*. 2011;16(2):180–187.
- O'Callaghan JM, Knight SR, Morgan RD, Morris PJ. Preservation solutions for static cold storage of kidney allografts: a systematic review and meta-analysis. *Am J Transplant*. 2012;12(4):896–906.
- Muhlbacher F, Langer F, Mittermayer C. Preservation solutions for transplantation. *Transplant Proc*. 1999;31(5):2069–2070.
- Timsit MO, Tullius SG. Hypothermic kidney preservation: a remembrance of the past in the future? *Curr Opin Organ Transplant*. 2011;16(2):162–168.
- Carrel A, Lindbergh CA. The Culture of Whole Organs. *Science*. 1935;81(2112):621–623.
- Belzer FO, Ashby BS, Gulyassy PF, Powell M. Successful seventeen-hour preservation and transplantation of human-cadaver kidney. *N Engl J Med*. 1968;278(11):608–610.
- Collins GM, Bravo-Shugarman M, Terasaki PI. Kidney preservation for transportation. Initial perfusion and 30 hours' ice storage. *Lancet*. 1969;2(7632):1219–1222.
- Dreikorn K, Horsch R, Rohl L. 48- to 96-hour preservation of canine kidneys by initial perfusion and hypothermic storage using the Euro-Collins solution. *Eur Urol*. 1980;6(4):221–224.
- Ross H, Marshall VC, Escott ML. 72-hr canine kidney preservation without continuous perfusion. *Transplantation*. 1976;21(6):498–501.
- O'Callaghan JM, Knight SR, Morgan RD, Morris PJ. A National Registry Analysis of Kidney Allografts Preserved With Marshall's Solution in the United Kingdom. *Transplantation*. 2016;100(11):2447–2452.
- Wahlberg JA, Southard JH, Belzer FO. Development of a cold storage solution for pancreas preservation. *Cryobiology*. 1986;23(6):477–482.
- Bretschneider HJ. Myocardial protection. *Thorac Cardiovasc Surg*. 1980;28(5):295–302.
- Bellamy CA, Nicely B, Mattice BJ, Teaster R. Comparative analysis of clinical efficacy and cost between University of Wisconsin solution and histidine-tryptophan-ketoglutarate. *Prog Transplant*. 2008;18(3):166–171; quiz 172.
- Menasche P, Termignon JL, Pradier F, Grousset C, Mouas C, Alberici G, Weiss M, Piwnica A, Bloch G. Experimental evaluation of Celsior, a new heart preservation solution. *Eur J Cardiothorac Surg*. 1994;8(4):207–213.
- Collins GM, Bravo-Shugarman M, Novom S, Terasaki PI. Kidney preservation for transplantation. I. Twelve-hour storage in rabbits. *Transplant Proc*. 1969;1(3):801–807.
- Ben Abdennebi H, Steghens JP, Hadj-Aissa A, Barbieux A, Ramella-Virieux S, Gharib C, Boillot O. A preservation solution with polyethylene glycol and calcium: a possible multi-organ liquid. *Transpl Int*. 2002;15(7):348–354.
- de Boer J, De Meester J, Smits JM, Groenewoud AF, Bok A, van der Velde O, Doxiadis II, Persijn GG. Eurotransplant randomized multicenter kidney graft preservation study comparing HTK with UW and Euro-Collins. *Transpl Int*. 1999;12(6):447–453.

25. Erhard J, Lange R, Scherer R, Kox WJ, Bretschneider HJ, Gebhard MM, Eigler FW. Comparison of histidine-tryptophan-ketoglutarate (HTK) solution versus University of Wisconsin (UW) solution for organ preservation in human liver transplantation. A prospective, randomized study. *Transpl Int*. 1994;7(3):177–181.
26. Voigt MR, DeLario GT. Perspectives on abdominal organ preservation solutions: a comparative literature review. *Prog Transplant*. 2013;23(4):383–391.
27. Southard JH, van Gulik TM, Ametani MS, Vreugdenhil PK, Lindell SL, Pienaar BL, Belzer FO. Important components of the UW solution. *Transplantation*. 1990;49(2):251–257.
28. Belzer FO, Sollinger HW, Glass NR, Miller DT, Hoffmann RM, Southard JH. Beneficial effects of adenosine and phosphate in kidney preservation. *Transplantation*. 1983;36(6):633–635.
29. Ploeg RJ, Goossens D, McAnulty JF, Southard JH, Belzer FO. Successful 72-hour cold storage of dog kidneys with UW solution. *Transplantation*. 1988;46(2):191–196.
30. Ploeg RJ. Kidney preservation with the UW and Euro-Collins solutions. A preliminary report of a clinical comparison. *Transplantation*. 1990;49(2):281–284.
31. Alshabani K, Nizamuddin N, Raza S, Alfurayh O, Almehari K, Qunibi W, Sanjad E. University of Wisconsin versus Eurocollins solution for kidney preservation: analysis of clinical outcome. *Transplant Proc*. 1998;30(7):3681–3682.
32. Tojimbara T, Wicomb WN, Garcia-Kennedy R, Burns W, Hayashi M, Collins G, Esquivel CO. Liver transplantation from non-heart beating donors in rats: influence of viscosity and temperature of initial flushing solutions on graft function. *Liver Transpl Surg*. 1997;3(1):39–45.
33. Shahed A, Tarin T, Jones EA, Shoskes DA. Comparison of preservation solution RPS-96 with University of Wisconsin solution in rat renal ischemia-reperfusion injury. *Transplant Proc*. 2000;32(4):806–807.
34. Candinas D, Largiader F, Binswanger U, Sutherland DE, Schlumpf R. A novel dextran 40-based preservation solution. *Transpl Int*. 1996;9(1):32–37.
35. Maluf DG, Mas VR, Yanek K, Stone JJ, Weis R, Massey D, Spiess B, Posner MP, Fisher RA. Molecular markers in stored kidneys using perfluorocarbon-based preservation solution: preliminary results. *Transplant Proc*. 2006;38(5):1243–1246.
36. Li S, Constantinescu I, Guan Q, Kalathottukaren MT, Brooks DE, Nguan CY, Kizhakkedathu JN, Du C. Advantages of replacing hydroxyethyl starch in University of Wisconsin solution with hyperbranched polyglycerol for cold kidney perfusion. *J Surg Res*. 2016;205(1):59–69.
37. Zhu Y, Furukawa H, Nakamura K, Hamamoto I, Wu Y, Xiaoshan R, Venkataramanan R, Todo S, Starzl TE. Sodium lactobionate sucrose solution for canine liver and kidney preservation. *Transplant Proc*. 1993;25(1 Pt 2):1618–1619.
38. Gubernatis G, Pichlmayr R, Lamesch P, Grosse H, Bornscheuer A, Meyer HJ, Ringe B, Farle M, Bretschneider HJ. HTK-solution (Bretschneider) for human liver transplantation. First clinical experiences. *Langenbecks Arch Chir*. 1990;375(2):66–70.
39. Lindell SL, Gandolph D, Southard JH, Belzer FO. Comparison of PBS, HTK, and UW solutions for kidney preservation. *Transplant Proc*. 1991;23(5):2399–2401.
40. Groenewoud AF, Thorogood J. Current status of the Eurotransplant randomized multicenter study comparing kidney graft preservation with histidine-tryptophan-ketoglutarate, University of Wisconsin, and Euro-Collins solutions. The HTK Study Group. *Transplant Proc*. 1993;25(1 Pt 2):1582–1585.
41. de Boer J, Smits JM, De Meester J, van der Velde O, Bok A, Persijn GG, Ringe B. A randomized multicenter study on kidney preservation comparing HTK with UW. *Transplant Proc*. 1999;31(5):2065–2066.
42. Trushkov S, Bicans J, Shevelev V, Jushinskis J, Suhorukov V, Rozental R. Use of HTK solution in kidney preservation. *Transplant Proc*. 2003;35(2):766.
43. Stewart ZA, Cameron AM, Singer AL, Montgomery RA, Segev DL. Histidine-Tryptophan-Ketoglutarate (HTK) is associated with reduced graft survival in deceased donor livers, especially those donated after cardiac death. *Am J Transplant*. 2009;9(2):286–293.
44. Karam G, Compagnon P, Hourmant M, Despins P, Duveau D, Noury D, Boudjema K. A single solution for multiple organ procurement and preservation. *Transpl Int*. 2005;18(6):657–663.
45. Jovine E, Di Benedetto F, Quintini C, Masetti M, Cautero N, Gelmini R, Andreotti A, Bezer L, Sassi S, Boggi U, Filipponi F, et al. Procurement technique for isolated small bowel, pancreas, and liver from multiorgan cadaveric donor. *Transplant Proc*. 2002;34(3):904–905.
46. Faenza A, Catena F, Nardo B, Montalti R, Capocasale E, Busi N, Boggi U, Vistoli F, Di Naro A, Albertazzi A, Mosca F, et al. Kidney preservation with university of Wisconsin and Celsior solution: a prospective multicenter randomized study. *Transplantation*. 2001;72(7):1274–1277.
47. Perez Sanz P, Burgos Revilla FJ, Marcen Letosa R, Pascual Santos J, Merino Rivas JL, Ortuno Mirete J. [Celsior's kidney preservation in renal transplantation. Our experience]. *Actas Urol Esp*. 2004;28(1):49–53.
48. Liu SY. [Preparation of a new kidney perfusate HC-A (hypertonic citrate adenine solution) and its clinical application]. *Zhonghua Wai Ke Za Zhi*. 1982;20(7):438–439.
49. Sui M, Zhang L, Yang J, Zeng L, Zhao W, Zhu Y. A new HC-A II solution for kidney preservation: a multi-center randomized controlled trial in China. *Ann Transplant*. 2014;19:614–620.
50. Zhu Y, Zhang L, Min Z, He C. The Thirty-Six Year Experience in Kidney Transplantation at Changzheng Hospital. *Clin Transpl*. 2014:77–81.
51. Ahmad N, Pratt JR, Potts DJ, Lodge JP. Comparative efficacy of renal preservation solutions to limit functional impairment after warm ischemic injury. *Kidney Int*. 2006;69(5):884–893.
52. Jamieson RW, Friend PJ. PBS140: new competition in the organ preservation market? *Kidney Int*. 2006;69(5):784–785.
53. Norby J, Jacobsen IA, Pegg DE, Starklint H, Chemnitz J, Diaper MP. Preservation of rabbit kidneys using a solution containing hydrolyzed starch. *Transplantation*. 1991;52(5):799–804.

54. Lopez-Neblina F, Toledo AH, Toledo-Pereyra LH. Evaluation of a novel cold storage solution (HBs) in a rat kidney transplant model. *J Invest Surg.* 2007;20(4):257–263.
55. Fehrenberg C, von Baeyer H, Unger V, Schmitt R, Haider W, Quarcoo D, Groneberg DA, Grosse-Siestrup C. Protective effects of B2 preservation solution in comparison to a standard solution (histidine-tryptophan-ketoglutarate/Bretschneider) in a model of isolated autologous hemoperfused porcine kidney. *Nephron Physiol.* 2004;96(2): p52–58.
56. Regner KR, Nilakantan V, Ryan RP, Mortensen J, White SM, Shames BD, Roman RJ. Protective effect of Lifor solution in experimental renal ischemia-reperfusion injury. *J Surg Res.* 2010;164(2): e291–297.
57. Gage F, Leeser DB, Porterfield NK, Graybill JC, Gillern S, Hawksworth JS, Jindal RM, Thai N, Falta EM, Tadaki DK, Brown TS, et al. Room temperature pulsatile perfusion of renal allografts with Lifor compared with hypothermic machine pump solution. *Transplant Proc.* 2009;41(9):3571–3574.
58. Kalenski J, Mancina E, Paschenda P, Beckers C, Bleilevens C, Tothova L, Boor P, Doorschodt BM, Tolba RH. Improved preservation of warm ischemia-damaged porcine kidneys after cold storage in Ecosol, a novel preservation solution. *Ann Transplant.* 2015;20:233–242.
59. Cierpka L, Ryszka F, Dolinska B, Smorag Z, Slomski R, Wia-derkiewicz R, Caban A, Budzinski G, Oczkowicz G, Wiczorek J. Biolasol: novel perfusion and preservation solution for kidneys. *Transplant Proc.* 2014;46(8):2539–2541.
60. Dolinska B, Ostrozka-Cieslik A, Caban A, Cierpka L, Ryszka F. Comparing the effect of Biolasol(R) and HTK solutions on maintaining proper homeostasis, indicating the kidney storage efficiency prior to transplantation. *Ann Transplant.* 2012;17(2):74–78.
61. Khirabadi BS, Fahy GM. Cryopreservation of the mammalian kidney. I. Transplantation of rabbit kidneys perfused with EC and RPS-2 at 2–4 degrees C. *Cryobiology.* 1994;31(1): 10–25.
62. Herrero I, Torras J, Carrera M, Castells A, Pasto L, Gil-Vernet S, Alsina J, Grinyo JM. Evaluation of a preservation solution containing fructose-1,6-diphosphate and mannitol using the isolated perfused rat kidney. Comparison with Euro-Collins and University of Wisconsin solutions. *Nephrol Dial Transplant.* 1995;10(4):519–526.
63. Kay MD, Hosgood SA, Harper SJ, Bagul A, Waller HL, Rees D, Nicholson ML. Static normothermic preservation of renal allografts using a novel nonphosphate buffered preservation solution. *Transpl Int.* 2007;20(1):88–92.
64. Wang S, Gumpfer K, Tan T, Luo X, Guo H, Ming C, Jiang H, Fang J, Du G, Zhu H, Ma J, et al. A novel organ preservation solution with efficient clearance of red blood cells improves kidney transplantation in a canine model. *Cell Biosci.* 2018;8:28.
65. Zheng JH, Min ZL, Li YL, Zhu YH, Ye TJ, Li JQ, Pan TW, Ding GS, Wang ML. A modified CZ-1 preserving solution for organ transplantation: comparative study with UW preserving solution. *Chin Med J (Engl).* 2008;121(10):904–909.
66. Ahmad N, Hostert L, Pratt JR, Billar KJ, Potts DJ, Lodge JP. A pathophysiologic study of the kidney tubule to optimize organ preservation solutions. *Kidney Int.* 2004;66(1):77–90.
67. Wilson CH, Brook NR, Talbot D. Preservation solutions for solid organ transplantation. *Mini Rev Med Chem.* 2006;6(10): 1081–1090.
68. Badet L, Ben Abdennebi H, Petruzzo P, McGregor B, Espa M, Hadj-Aissa A, Ramella-Virieux S, Steghens JP, Portoghesse F, Martin X. Effect of IGL-1, a new preservation solution, on kidney grafts (a pre-clinical study). *Transpl Int.* 2005;17(12): 815–821.
69. Badet L, Abdennebi HB, Petruzzo P, McGregor B, Espa M, Hadj-Aissa A, Ramella-Virieux S, Steghens JP, Portoghesse F, Morelon E, Martin X. [Evaluation of IGL-1, a new organ preservation solution: preclinical results in renal transplantation]. *Prog Urol.* 2005;15(3):481–488; discussion 487.
70. Katayama M, Tsuchiaka S, Motegi T, Miyazaki M, Yamashita T, Shimamura S, Okamura Y, Uzuka Y. High-molecular-weight polyethylene glycol enhances hypothermic storage of feline kidney cells. *J Vet Med Sci.* 2014;76(6):923–926.
71. Fuller BJ, Shurey C, Lane N, Petrenko A, Green C. Hypothermic renal preservation with a sucrose/ polyethylene glycol solution in a rabbit renal transplant model. *Cryo Letters.* 2006;27(2):127–132.
72. Faure JP, Hauet T, Han Z, Goujon JM, Petit I, Mauco G, Eugene M, Carretier M, Papadopoulos V. Polyethylene glycol reduces early and long-term cold ischemia-reperfusion and renal medulla injury. *J Pharmacol Exp Ther.* 2002;302(3): 861–870.
73. Hauet T, Baumert H, Amor IB, Goujon JM, Gibelin H, Godart C, Vandewalle A, Carretier M, Eugene M. Protection of auto-transplanted pig kidneys from ischemia-reperfusion injury by polyethylene glycol. *Transplantation.* 2000;70(11):1569–1575.
74. Hauet T, Goujon JM, Baumert H, Petit I, Carretier M, Eugene M, Vandewalle A. Polyethylene glycol reduces the inflammatory injury due to cold ischemia/reperfusion in autotransplanted pig kidneys. *Kidney Int.* 2002;62(2):654–667.
75. Gallinat A, Luer B, Swoboda S, Rauen U, Paul A, Minor T. Use of the new preservation solution Custodiol-N supplemented with dextran for hypothermic machine perfusion of the kidney. *Cryobiology.* 2013;66(2):131–135.
76. Lobb I, Davison M, Carter D, Liu W, Haig A, Gunaratnam L, Sener A. Hydrogen sulfide treatment mitigates renal allograft ischemia-reperfusion injury during cold storage and improves early transplant kidney function and survival following allogeneic renal transplantation. *J Urol.* 2015;194(6):1806–1815.
77. Lobb I, Mok A, Lan Z, Liu W, Garcia B, Sener A. Supplemental hydrogen sulphide protects transplant kidney function and prolongs recipient survival after prolonged cold ischaemia-reperfusion injury by mitigating renal graft apoptosis and inflammation. *BJU Int.* 2012;110(11 Pt C): E1187–1195.
78. Abe T, Li XK, Yazawa K, Hatayama N, Xie L, Sato B, Kakuta Y, Tsutahara K, Okumi M, Tsuda H, Kaimori JY, et al. Hydrogen-rich University of Wisconsin solution attenuates renal cold ischemia-reperfusion injury. *Transplantation.* 2012;94(1):14–21.

79. Ozaki KS, Yoshida J, Ueki S, Pettigrew GL, Ghonem N, Sico RM, Lee LY, Shapiro R, Lakkis FG, Pacheco-Silva A, Murase N. Carbon monoxide inhibits apoptosis during cold storage and protects kidney grafts donated after cardiac death. *Transpl Int*. 2012;25(1):107–117.
80. Nakao A, Faleo G, Shimizu H, Nakahira K, Kohmoto J, Sugimoto R, Choi AM, McCurry KR, Takahashi T, Murase N. Ex vivo carbon monoxide prevents cytochrome P450 degradation and ischemia/reperfusion injury of kidney grafts. *Kidney Int*. 2008;74(8):1009–1016.
81. Yoshida J, Ozaki KS, Nalesnik MA, Ueki S, Castillo-Rama M, Faleo G, Ezzelarab M, Nakao A, Eksler B, Echeverri GJ, Ross MA, et al. Ex vivo application of carbon monoxide in UW solution prevents transplant-induced renal ischemia/reperfusion injury in pigs. *Am J Transplant*. 2010;10(4):763–772.
82. Sener A, Tran KC, Deng JP, Garcia B, Lan Z, Liu W, Sun T, Arp J, Salna M, Acott P, Cepinskas G, et al. Carbon monoxide releasing molecules inhibit cell death resulting from renal transplantation related stress. *J Urol*. 2013;190(2):772–778.
83. Thuillier R, Dutheil D, Trieu MT, Mallet V, Allain G, Rouselot M, Denizot M, Goujon JM, Zal F, Hauet T. Supplementation with a new therapeutic oxygen carrier reduces chronic fibrosis and organ dysfunction in kidney static preservation. *Am J Transplant*. 2011;11(9):1845–1860.
84. Faure A, Bruzzese L, Steinberg JG, Jammes Y, Torrents J, Berdah SV, Garnier E, Legris T, Loundou A, Chalopin M, Magalon G, et al. Effectiveness of pure argon for renal transplant preservation in a preclinical pig model of heterotopic autotransplantation. *J Transl Med*. 2016;14:40.
85. Inman SR, Burns TE, Plott WK, Pomilee RA, Antonelli JA, Lewis RM. Endothelin receptor blockade during hypothermic perfusion preservation mitigates the adverse effect of pre-retrieval warm ischemic injury on posttransplant glomerular filtration rate. *Transplantation*. 2002;74(2):164–168.
86. Hammad FT, Davis G, Zhang XY, Wheatley AM. Endothelin ETA and ETB receptor antagonism during cold preservation in renal transplantation. *Transplantation*. 2001;71(5):619–627.
87. Hammad FT, Davis G, Zhang X, Wheatley AM. The role of endothelin in early renal cortical reperfusion in renal transplantation. *Eur Surg Res*. 2000;32(6):380–388.
88. Inman SR, Burns TE, Plott WK, Pomilee RA, Antonelli JA, Lewis RM. Addition of an endothelin receptor antagonist to modified Belzer's perfusion preservation solution mitigates the adverse effect of pre-retrieval warm ischemic injury on post-transplant glomerular filtration rate. *Transplant Proc*. 2001;33(6):2975–2976.
89. Kirchner C, Luer B, Efferz P, Wohlschlaeger J, Paul A, Minor T. Ex vivo use of a Rho-kinase inhibitor during renal preservation improves graft function upon reperfusion. *Cryobiology*. 2015;70(1):71–75.
90. Giraud S, Thuillier R, Belliard A, Hebrard W, Nadeau C, Milin S, Goujon JM, Manguy E, Mauco G, Hauet T, Macchi L. Direct thrombin inhibitor prevents delayed graft function in a porcine model of renal transplantation. *Transplantation*. 2009;87(11):1636–1644.
91. Favreau F, Thuillier R, Cau J, Milin S, Manguy E, Mauco G, Zhu X, Lerman LO, Hauet T. Anti-thrombin therapy during warm ischemia and cold preservation prevents chronic kidney graft fibrosis in a DCD model. *Am J Transplant*. 2010;10(1):30–39.
92. Thuillier R, Favreau F, Celhay O, Macchi L, Milin S, Hauet T. Thrombin inhibition during kidney ischemia-reperfusion reduces chronic graft inflammation and tubular atrophy. *Transplantation*. 2010;90(6):612–621.
93. McAnulty JF, Reid TW, Waller KR, Murphy CJ. Successful six-day kidney preservation using trophic factor supplemented media and simple cold storage. *Am J Transplant*. 2002;2(8):712–718.
94. Desurmont T, Giraud S, Cau J, Goujon JM, Scepti M, Roumy J, Chatauret N, Thuillier R, Hauet T. Trophic factor and FR167653 supplementation during cold storage rescue chronic renal injury. *J Urol*. 2011;185(3):1139–1146.
95. Nakatani T, Kim T, Uchida J, Kumata N, Kawashima H, Sugimura K. Hepatocyte growth factor ameliorates renal hemodynamic disorder after ischemia/reperfusion. *Int J Mol Med*. 2002;10(2):217–219.
96. Petrinc D, Reilly JM, Sicard GA, Lowell JA, Howard TK, Martin DR, Brennan DC, Miller SB. Insulin-like growth factor-I attenuates delayed graft function in a canine renal autotransplantation model. *Surgery*. 1996;120(2):221–225; discussion 225–226.
97. McAnulty JF, Foley JD, Reid TW, Heath TD, Waller KR, Murphy CJ. Suppression of cold ischemic injury in stored kidneys by the antimicrobial peptide bactenecin. *Cryobiology*. 2004;49(3):230–240.
98. Celic T, Spanjol J, Grskovic A, Markic D, Prebilib I, Fuckar Z, Bobinac D. Bone morphogenetic protein-7 reduces cold ischemic injury in rat kidney. *Transplant Proc*. 2011;43(7):2505–2509.
99. Ramackers W, Klose J, Tiede A, Werwitzke S, Rataj D, Friedrich L, Johanning K, Vondran FW, Bergmann S, Schuettler W, Bockmeyer CL, et al. Effect of TNF-alpha blockade on coagulopathy and endothelial cell activation in xenoperfused porcine kidneys. *Xenotransplantation*. 2015;22(4):284–294.
100. Yang C, Hosgood SA, Meeta P, Long Y, Zhu T, Nicholson ML, Yang B. Cyclic Helix B Peptide in Preservation Solution and Autologous Blood Perfusate Ameliorates Ischemia-Reperfusion Injury in Isolated Porcine Kidneys. *Transplant Direct*. 2015;1(2):e6.
101. Erkasap S, Ates E. L-Arginine-enriched preservation solution decreases ischaemia/reperfusion injury in canine kidneys after long-term cold storage. *Nephrol Dial Transplant*. 2000;15(8):1224–1227.
102. Yang B, Hosgood SA, Nicholson ML. Naked small interfering RNA of caspase-3 in preservation solution and autologous blood perfusate protects isolated ischemic porcine kidneys. *Transplantation*. 2011;91(5):501–507.
103. Moser MA, Arcand S, Lin HB, Wojnarowicz C, Sawicka J, Banerjee T, Luo Y, Beck GR, Luke PP, Sawicki G. Protection of the transplant kidney from preservation injury by inhibition

- of matrix metalloproteinases. *PLoS One*. 2016;11(6): e0157508.
104. Chen W, Bennett CF, Wang ME, Dragun D, Tian L, Stecker K, Clark JH, Kahan BD, Stepkowski SM. Perfusion of kidneys with unformulated "naked" intercellular adhesion molecule-1 antisense oligodeoxynucleotides prevents ischemic/reperfusion injury. *Transplantation*. 1999;68(6): 880–887.
105. Wingenfeld P, Gehrman U, Strubind S, Minor T, Isselhard W, Michalk DV. Long-lasting hypoxic preservation of porcine kidney cells. Beneficial effect of taurine on viability and metabolism in a simplified transplantation model. *Adv Exp Med Biol*. 1996;403:203–212.
106. Wingenfeld P, Strubind S, Gehrman U, Minor T, Isselhard W, Michalk D. Protecting effect of taurine against hypoxic cell damage in renal tubular cells cultured in different transplant preservation solutions. *Adv Exp Med Biol*. 1994;359: 159–169.
107. Lodge JP, Lam FT, Perry SL, Giles GR. Ranolazine—a new drug with beneficial effects on renal preservation. *Transplantation*. 1990;50(5):755–759.
108. Nakamoto M, Shapiro JI, Mills SD, Schrier RW, Chan L. Improvement of renal preservation by verapamil with 24-hour cold perfusion in the isolated rat kidney. *Transplantation*. 1988;45(2):313–315.
109. Shapiro JI, Cheung C, Itabashi A, Chan L, Schrier RW. The effect of verapamil on renal function after warm and cold ischemia in the isolated perfused rat kidney. *Transplantation*. 1985;40(6):596–600.
110. Tucci S, Jr., Borelli-Bovo TJ, Cologna AJ, Tiraboschi RB, Martins AC, Roselino JE. Calcium channel blocker and renal mitochondrial function in warm renal ischemia. *Int Braz J Urol*. 2005;31(4):384–389.
111. Baumert H, Faure JP, Zhang K, Petit I, Goujon JM, Dutheil D, Favreau F, Barriere M, Tillement JP, Mauco G, Papadopoulos V, et al. Evidence for a mitochondrial impact of trimetazidine during cold ischemia and reperfusion. *Pharmacology*. 2004; 71(1):25–37.
112. Faure JP, Baumert H, Han Z, Goujon JM, Favreau F, Dutheil D, Petit I, Barriere M, Tallineau C, Tillement JP, Carretier M, et al. Evidence for a protective role of trimetazidine during cold ischemia: targeting inflammation and nephron mass. *Biochem Pharmacol*. 2003;66(11):2241–2250.
113. Goujon JM, Vandewalle A, Baumert H, Carretier M, Hauet T. Influence of cold-storage conditions on renal function of auto-transplanted large pig kidneys. *Kidney Int*. 2000;58(2): 838–850.
114. Hauet T, Goujon JM, Vandewalle A, Baumert H, Lacoste L, Tillement JP, Eugene M, Carretier M. Trimetazidine reduces renal dysfunction by limiting the cold ischemia/reperfusion injury in autotransplanted pig kidneys. *J Am Soc Nephrol*. 2000;11(1):138–148.
115. Hauet T, Mothes D, Goujon J, Germonville T, Caritez JC, Carretier M, Eugene M, Tillement J. Trimetazidine reverses deleterious effects of ischemia-reperfusion in the isolated perfused pig kidney model. *Nephron*. 1998;80(3):296–304.
116. Hauet T, Tallineau C, Goujon JM, Carretier M, Eugene M, Tillement JP. Efficiency of trimetazidine in renal dysfunction secondary to cold ischemia-reperfusion injury: a proposed addition to University of Wisconsin solution. *Cryobiology*. 1998;37(3):231–244.
117. McAnulty JF, Huang XQ. The efficacy of antioxidants administered during low temperature storage of warm ischemic kidney tissue slices. *Cryobiology*. 1997;34(4):406–415.
118. Nakagawa K, Koo DD, Davies DR, Gray DW, McLaren AJ, Welsh KI, Morris PJ, Fuggle SV. Lecithinized superoxide dismutase reduces cold ischemia-induced chronic allograft dysfunction. *Kidney Int*. 2002;61(3):1160–1169.
119. Tahara M, Nakayama M, Jin MB, Fujita M, Suzuki T, Taniguchi M, Shimamura T, Furukawa H, Todo S. A radical scavenger, edaravone, protects canine kidneys from ischemia-reperfusion injury after 72 hours of cold preservation and auto-transplantation. *Transplantation*. 2005;80(2):213–221.
120. Mitchell T, Rotaru D, Saba H, Smith RA, Murphy MP, MacMillan-Crow LA. The mitochondria-targeted antioxidant mitoquinone protects against cold storage injury of renal tubular cells and rat kidneys. *J Pharmacol Exp Ther*. 2011; 336(3):682–692.
121. Parajuli N, Campbell LH, Marine A, Brockbank KG, Macmillan-Crow LA. MitoQ blunts mitochondrial and renal damage during cold preservation of porcine kidneys. *PLoS One*. 2012;7(11): e48590.
122. McAnulty JF, Huang XQ. Nonesterified fatty acid content of rabbit kidney cortex slices after warm ischemia and low temperature preservation. *Cryobiology*. 1996;33(1):70–81.
123. Dobashi K, Singh I, Orak JK, Asayama K, Singh AK. Combination therapy of N-acetylcysteine, sodium nitroprusside and phosphoramidon attenuates ischemia-reperfusion injury in rat kidney. *Mol Cell Biochem*. 2002;240(1–2):9–17.
124. Treska V, Kuntscher V, Molacek J, Kobr J, Racek J, Trefil L. Can the ischemia-reperfusion syndrome in transplanted kidneys procured from non-heart-beating donors be influenced by adding selenium into the reperfusion solution? An experimental study. *Transplant Proc*. 2003;35(4):1584–1586.
125. Treska V, Kuntscher V, Molacek J, Kobr J, Racek J, Trefil L. Can ischemia-reperfusion syndrome in transplanted kidneys procured from non-heart-beating donors be influenced by adding selenium into the reperfusion solution? An experimental study. *Transplant Proc*. 2003;35(8):3125–3127.
126. Masaki Y, Kumano K, Endo T, Iwamura M, Koshiba K, Yokota K, Okubo M. Protective effect of nicaraven against prolonged cold kidney preservation and reperfusion injury. *Transplant Proc*. 1998;30(7):3758–3760.
127. Snoeijs MG, Vaahtera L, de Vries EE, Schurink GW, Haenen GR, Peutz-Kootstra CJ, Buurman WA, van Heurn LW, Parkkinen J. Addition of a water-soluble propofol formulation to preservation solution in experimental kidney transplantation. *Transplantation*. 2011;92(3):296–302.
128. Polyak MM, Arrington BO, Stubenbord WT, Kapur S, Kinkhabwala M. Prostaglandin E1 influences pulsatile preservation characteristics and early graft function in expanded criteria donor kidneys. *J Surg Res*. 1999;85(1):17–25.

129. Polyak MM, Arrington BO, Stubenbord WT, Kinkhabwala M. Prostaglandin E1 improves pulsatile preservation characteristics and early graft function in expanded criteria donor kidneys. *ASAIO J*. 1998;44(5):M610–M612.
130. Zhang X, He D, Xu L, Ling S. Protective effect of tanshinone IIA on rat kidneys during hypothermic preservation. *Mol Med Rep*. 2012;5(2):405–409.
131. Belzer FO, Southard JH. Principles of solid-organ preservation by cold storage. *Transplantation*. 1988;45(4):673–676.
132. Hoffmann RM, Southard JH, Lutz M, Mackety A, Belzer FO. Synthetic perfusate for kidney preservation. Its use in 72-hour preservation of dog kidneys. *Arch Surg*. 1983;118(8):919–921.
133. Schlumpf R, Morel P, Loveras JJ, Condie RM, Matas A, Kurlle J, Fasola CG, Najarian JS, Sutherland DE. Dextran 40 successfully replaces the non-essential hydroxyethylstarch in the University of Wisconsin solution for 72-hour simple cold storage of the canine kidney. *Transplant Proc*. 1991;23(1 Pt 1):657–659.
134. Hauet T, Eugene M. A new approach in organ preservation: potential role of new polymers. *Kidney Int*. 2008;74(8):998–1003.
135. Thuillier R, Renard C, Rogel-Gaillard C, Demars J, Milan D, Forestier L, Ouldoulene A, Goujon JM, Badet L, Hauet T. Effect of polyethylene glycol-based preservation solutions on graft injury in experimental kidney transplantation. *Br J Surg*. 2011;98(3):368–378.
136. Maathuis MH, Ottens PJ, van Goor H, Zwaagstra JJ, Wiersma-Buist J, Schuurs TA, Ploeg RJ, Leuvenink HG. Static cold storage preservation of ischemically damaged kidneys. a comparison between IGL-1 and UW solution. *Transpl Int*. 2008;21(5):473–482.
137. Cudas R, Petruzzo P, Morelon E, Lefrancois N, Danjou F, Berthillot C, Contu P, Espa M, Martin X, Badet L. IGL-1 solution in kidney transplantation: first multi-center study. *Clin Transplant*. 2009;23(3):337–342.
138. Bessems M, Doorschodt BM, van Vliet AK, van Gulik TM. Machine perfusion preservation of the non-heart-beating donor rat livers using polysol, a new preservation solution. *Transplant Proc*. 2005;37(1):326–328.
139. Schreinemachers MC, Doorschodt BM, Florquin S, van den Bergh Weerman MA, Reitsma JB, Lai W, Sitzia M, Minor TM, Tolba RH, van Gulik TM. Improved preservation and microcirculation with POLYSOL after transplantation in a porcine kidney autotransplantation model. *Nephrol Dial Transplant*. 2009;24(3):816–824.
140. Schreinemachers MC, Doorschodt BM, Florquin S, Tolba RH. Comparison of preservation solutions for washout of kidney grafts: an experimental study. *Transplant Proc*. 2009;41(10):4072–4079.
141. Schreinemachers MC, Doorschodt BM, Florquin S, Idu MM, Tolba RH, van Gulik TM. Improved renal function of warm ischemically damaged kidneys using Polysol. *Transplant Proc*. 2009;41(1):32–35.
142. Schreinemachers MC, Bemelman FJ, Idu MM, van Donselaar-van der Pant KA, van de Berg PJ, Reitsma JB, Legemate DA, Florquin S, ten Berge IJ, Doorschodt BM, van Gulik TM. First clinical experience with polysol solution: pilot study in living kidney transplantation. *Transplant Proc*. 2013;45(1):38–45.
143. Giraud S, Hauet T, Eugene M, Mauco G, Barrou B. A new preservation solution (SCOT 15) Improves the islet isolation process from pancreata of non-heart-beating donors: a Murine model. *Transplant Proc*. 2009;41(8):3293–3295.
144. Billault C, Vaessen C, Van Glabeke E, Rolland E, Ourahma S, Dimitru L, Richard F, Eugene M, Barrou B. Use of the SCOT solution in kidney transplantation: preliminary report. *Transplant Proc*. 2006;38(7):2281–2282.
145. Dutheil D, Underhaug Gjerde A, Petit-Paris I, Mauco G, Holmsen H. Polyethylene glycols interact with membrane glycerophospholipids: is this part of their mechanism for hypothermic graft protection? *J Chem Biol*. 2009;2(1):39–49.
146. Liu G, Fu L, Zhang G. Role of hydrophobic interactions in the adsorption of poly(ethylene glycol) chains on phospholipid membranes investigated with a quartz crystal microbalance. *J Phys Chem B*. 2009;113(11):3365–3369.
147. Murad KL, Gosselin EJ, Eaton JW, Scott MD. Stealth cells: prevention of major histocompatibility complex class II-mediated T-cell activation by cell surface modification. *Blood*. 1999;94(6):2135–2141.
148. Moody BF, Calvert JW. Emergent role of gasotransmitters in ischemia-reperfusion injury. *Med Gas Res*. 2011;1(1):3.
149. Isselhard W, Berger M, Denecke H, Witte J, Fischer JH, Molzberger H. Metabolism of canine kidneys in anaerobic ischemia and in aerobic ischemia by persufflation with gaseous oxygen. *Pflugers Arch*. 1972;337(2):87–106.
150. Rolles K, Foreman J, Pegg DE. A pilot clinical study of retrograde oxygen persufflation in renal preservation. *Transplantation*. 1989;48(2):339–342.
151. Ohsawa I, Ishikawa M, Takahashi K, Watanabe M, Nishimaki K, Yamagata K, Katsura K, Katayama Y, Asoh S, Ohta S. Hydrogen acts as a therapeutic antioxidant by selectively reducing cytotoxic oxygen radicals. *Nat Med*. 2007;13(6):688–694.
152. Motterlini R, Otterbein LE. The therapeutic potential of carbon monoxide. *Nat Rev Drug Discov*. 2010;9(9):728–743.
153. Ozaki KS, Kimura S, Murase N. Use of carbon monoxide in minimizing ischemia/reperfusion injury in transplantation. *Transplant Rev (Orlando)*. 2012;26(2):125–139.
154. Lundberg JO, Weitzberg E, Gladwin MT. The nitrate-nitrite-nitric oxide pathway in physiology and therapeutics. *Nat Rev Drug Discov*. 2008;7(2):156–167.
155. Gladwin MT, Schechter AN. Nitric oxide therapy in sickle cell disease. *Semin Hematol*. 2001;38(4):333–342.
156. Szabo C. Hydrogen sulphide and its therapeutic potential. *Nat Rev Drug Discov*. 2007;6(11):917–935.
157. Elrod JW, Calvert JW, Morrison J, Doeller JE, Kraus DW, Tao L, Jiao X, Scalia R, Kiss L, Szabo C, Kimura H, et al. Hydrogen sulfide attenuates myocardial ischemia-reperfusion injury by preservation of mitochondrial function. *Proc Natl Acad Sci U S A*. 2007;104(39):15560–15565.

158. Shibuya N, Koike S, Tanaka M, Ishigami-Yuasa M, Kimura Y, Ogasawara Y, Fukui K, Nagahara N, Kimura H. A novel pathway for the production of hydrogen sulfide from D-cysteine in mammalian cells. *Nat Commun.* 2013;4:1366.
159. Ambiru S, Uryuhara K, Talpe S, Dehoux JP, Jacobbi L, Murphy CJ, McAnulty JF, Gianello P. Improved survival of orthotopic liver allograft in swine by addition of trophic factors to University of Wisconsin solution. *Transplantation.* 2004;77(2):302–319.
160. Kwon YS, Foley JD, Murphy CJ, McAnulty JF. The effect of trophic factor supplementation on cold ischemia-induced early apoptotic changes. *Transplantation.* 2007;83(1):91–94.
161. Riera M, Torras J, Herrero I, Valles J, Paubert-Braquet M, Cruzado JM, Alsina J, Grinyo JM. Neutrophils accentuate renal cold ischemia-reperfusion injury. Dose-dependent protective effect of a platelet-activating factor receptor antagonist. *J Pharmacol Exp Ther.* 1997;280(2):786–794.
162. Baker TA, Geng Q, Romero J, Picken MM, Gamelli RL, Majetschak M. Prolongation of myocardial viability by proteasome inhibition during hypothermic organ preservation. *Biochem Biophys Res Commun.* 2010;401(4):548–553.
163. Padrisa-Altes S, Zaouali MA, Bartrons R, Rosello-Catafau J. Ubiquitin-proteasome system inhibitors and AMPK regulation in hepatic cold ischaemia and reperfusion injury: possible mechanisms. *Clin Sci (Lond).* 2012;123(2):93–98.
164. Gao D, Ning N, Niu X, Hao G, Meng Z. Trimetazidine: a meta-analysis of randomised controlled trials in heart failure. *Heart.* 2011;97(4):278–286.
165. Zhao H, Alam A, Soo AP, George AJT, Ma D. Ischemia-reperfusion injury reduces long term renal graft survival: mechanism and beyond. *EBioMedicine.* 2018;28:31–42.
166. Eltzhig HK, Eckle T. Ischemia and reperfusion—from mechanism to translation. *Nat Med.* 2011;17(11):1391–1401.
167. Perico N, Cattaneo D, Sayegh MH, Remuzzi G. Delayed graft function in kidney transplantation. *Lancet.* 2004;364(9447):1814–1827.
168. Kosieradzki M, Rowinski W. Ischemia/reperfusion injury in kidney transplantation: mechanisms and prevention. *Transplant Proc.* 2008;40(10):3279–3288.
169. Dunbar LA, Caplan MJ. Ion pumps in polarized cells: sorting and regulation of the Na⁺, K⁺ and H⁺, K⁺-ATPases. *J Biol Chem.* 2001;276(32):29617–29620.
170. Hu LF, Li Y, Neo KL, Yong QC, Lee SW, Tan BK, Bian JS. Hydrogen sulfide regulates Na⁺/H⁺ exchanger activity via stimulation of phosphoinositide 3-kinase/Akt and protein kinase G pathways. *J Pharmacol Exp Ther.* 2011;339(2):726–735.
171. Patel KJ, Atkinson C, Broome A-M, Nadig SN. *Organ Preservation, Ischemia Reperfusion Injury, and Nanotherapeutics in Transplantation.* Technological Advances in Organ Transplantation. Cham: Springer; 2017:151–179.
172. Lam FT, Mavor AI, Potts DJ, Giles GR. Improved 72-hour renal preservation with phosphate-buffered sucrose. *Transplantation.* 1989;47(5):767–771.
173. Semenza GL. Life with oxygen. *Science.* 2007;318(5847):62–64.
174. Iwai Y, Kikuchi R, Watanabe Y, Koyama N, Hagiwara K, Nakamura H, Aoshiba K. *Extracellular Acidosis Inhibits Glucose Starvation-Induced Lung Cancer Cell Death by Suppressing Overall Metabolic Rate.* Philadelphia: AACR; 2018.
175. Baicu SC, Taylor MJ. Acid-base buffering in organ preservation solutions as a function of temperature: new parameters for comparing buffer capacity and efficiency. *Cryobiology.* 2002;45(1):33–48.
176. Malek M, Nematbakhsh M. Renal ischemia/reperfusion injury; from pathophysiology to treatment. *J Renal Inj Prev.* 2015;4(2):20–27.
177. Belous A, Knox C, Nicoud IB, Pierce J, Anderson C, Pinson CW, Chari RS. Altered ATP-dependent mitochondrial Ca²⁺ uptake in cold ischemia is attenuated by ruthenium red. *J Surg Res.* 2003;111(2):284–289.
178. Jassem W, Heaton ND. The role of mitochondria in ischemia/reperfusion injury in organ transplantation. *Kidney Int.* 2004;66(2):514–517.
179. Szczesny B, Modis K, Yanagi K, Coletta C, Le Trionnaire S, Perry A, Wood ME, Whiteman M, Szabo C. AP39, a novel mitochondria-targeted hydrogen sulfide donor, stimulates cellular bioenergetics, exerts cytoprotective effects and protects against the loss of mitochondrial DNA integrity in oxidatively stressed endothelial cells in vitro. *Nitric Oxide.* 2014;41:120–130.
180. Kosieradzki M, Kuczynska J, Piwowarska J, Wegrowicz-Rebandel I, Kwiatkowski A, Lisik W, Michalak G, Danielewicz R, Paczek L, Rowinski WA. Prognostic significance of free radicals: mediated injury occurring in the kidney donor. *Transplantation.* 2003;75(8):1221–1227.
181. Noiri E, Nakao A, Uchida K, Tsukahara H, Ohno M, Fujita T, Brodsky S, Goligorsky MS. Oxidative and nitrosative stress in acute renal ischemia. *Am J Physiol Renal Physiol.* 2018.
182. Ahlenstiel T, Burkhardt G, Kohler H, Kuhlmann MK. Improved cold preservation of kidney tubular cells by means of adding bioflavonoids to organ preservation solutions. *Transplantation.* 2006;81(2):231–239.
183. Dare AJ, Logan A, Prime TA, Rogatti S, Goddard M, Bolton EM, Bradley JA, Pettigrew GJ, Murphy MP, Saeb-Parsy K. The mitochondria-targeted anti-oxidant MitoQ decreases ischemia-reperfusion injury in a murine syngeneic heart transplant model. *J Heart Lung Transplant.* 2015;34(11):1471–1480.
184. Land W, Schneeberger H, Schleichner S, Illner WD, Abendroth D, Rutili G, Arfors KE, Messmer K. The beneficial effect of human recombinant superoxide dismutase on acute and chronic rejection events in recipients of cadaveric renal transplants. *Transplantation.* 1994;57(2):211–217.
185. Wang F, Yu G, Liu SY, Li JB, Wang JF, Bo LL, Qian LR, Sun XJ, Deng XM. Hydrogen-rich saline protects against renal ischemia/reperfusion injury in rats. *J Surg Res.* 2011;167(2):e339–344.
186. Gong W, Ma Y, Li A, Han S, Nie S. Trimetazidine suppresses oxidative stress, inhibits MMP-2 and MMP-9 expression, and prevents cardiac rupture in mice with myocardial infarction. *Cardiovascular Therapeutics.* 2018;36(5):e12460.

187. Kvietys PR, Granger DN. Role of reactive oxygen and nitrogen species in the vascular responses to inflammation. *Free Radic Biol Med.* 2012;52(3):556–592.
188. Haugen E, Nath KA. The involvement of oxidative stress in the progression of renal injury. *Blood Purif.* 1999;17(2–3): 58–65.
189. Chen GY, Nunez G. Sterile inflammation: sensing and reacting to damage. *Nat Rev Immunol.* 2010;10(12): 826–837.
190. Schroppel B, Legendre C. Delayed kidney graft function: from mechanism to translation. *Kidney Int.* 2014;86(2): 251–258.
191. Chin BY, Jiang G, Wegiel B, Wang HJ, Macdonald T, Zhang XC, Gallo D, Cszimadia E, Bach FH, Lee PJ, Otterbein LE. Hypoxia-inducible factor 1alpha stabilization by carbon monoxide results in cytoprotective preconditioning. *Proc Natl Acad Sci U S A.* 2007;104(12):5109–5114.
192. Hotchkiss RS, Strasser A, McDunn JE, Swanson PE. Cell death. *N Engl J Med.* 2009;361(16):1570–1583.
193. Green D. *Means to an End: Apoptosis and other Cell Death Mechanisms.* Cold Spring Harbor, NY: Cold Spring Harbor Laboratory Press; 2011.
194. Saelens X, Festjens N, Vande Walle L, van Gurp M, van Loo G, Vandenabeele P. Toxic proteins released from mitochondria in cell death. *Oncogene.* 2004;23(16):2861–2874.
195. Kumar S, Allen DA, Kieswich JE, Patel NS, Harwood S, Mazon E, Cuzzocrea S, Raftery MJ, Thiemermann C, Yaqoob MM. Dexamethasone ameliorates renal ischemia-reperfusion injury. *J Am Soc Nephrol.* 2009;20(11): 2412–2425.
196. Chan FK, Luz NF, Moriwaki K. Programmed necrosis in the cross talk of cell death and inflammation. *Annu Rev Immunol.* 2015;33:79–106.
197. Kang KJ. Mechanism of hepatic ischemia/reperfusion injury and protection against reperfusion injury. *Transplant Proc.* 2002;34(7):2659–2661.
198. Lempiainen J, Finckenberg P, Levijoki J, Mervaala E. AMPK activator AICAR ameliorates ischaemia reperfusion injury in the rat kidney. *Br J Pharmacol.* 2012;166(6):1905–1915.
199. Zhao W, Zhang L, Chen R, Lu H, Sui M, Zhu Y, Zeng L. SIRT3 protects against acute kidney injury via AMPK/mTOR-regulated autophagy. *Front Physiol.* 2018;9:1526.
200. Lee CY, Mangino MJ. Preservation methods for kidney and liver. *Organogenesis.* 2009;5(3):105–112.