Review Article

Preservation solutions for cardiac and pulmonary donor grafts: a review of the current literature

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Abstract: Hypothermic preservation of donor grafts is imperative to ameliorate ischemia related cellular damage prior to organ transplantation. Numerous solutions are in existence with widespread variability among transplant centers as to a consensus regarding the optimal preservation solution. Here, we present a concise review of pertinent preservation studies involving cardiac and pulmonary allografts in an attempt to minimize the variability among institutions and potentially improve graft and patient survival. A biochemical comparison of common preservation solutions was undertaken with an emphasis on Euro Collins (EC), University of Wisconsin (UW), histidine-tryptophan-ketoglutarate (HTK), Celsior (CEL), Perfadex (PER), Papworth, and Plegisol. An appraisal of the literature ensued containing the aforementioned preservation solutions in the setting of cardiac and pulmonary transplantation. Available evidence supports UW solution as the preservation solution of choice for cardiac transplants with encouraging outcomes relative to notable contenders such as CEL. Despite its success in the setting of cardiac transplantation, its use in pulmonary transplantation remains suboptimal and improved outcomes may be seen with PER. Together, we suggest, based on the literature that the use of UW solution and PER for cardiac and pulmonary transplants, respectively may improve transplant outcomes such as graft and patient survival.

Keywords: Preservation; donor; cardiac; pulmonary; transplantation

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Introduction

Despite many of the advances within the realm of transplantation, graft survival remains imperfect. Optimal preservation of the graft is an important determinant of graft survival and patient outcomes. Considerable attention is given to the *ex vivo* period as this segment represents a vulnerable timeframe whereby organs are susceptible to ongoing cellular damage that is further compounded by reperfusion injury upon re-anastomosis. Hypothermia is utilized to decrease the metabolic activity of donor organs during the *ex vivo* period. Decrease donor organ temperature from 37 to 4 ℃ results in a 12 fold decrease in the metabolic demand (1). However, hypothermia alone is unable to abolish all cellular damage as metabolism persists at approximately 5-10% of normal. In addition, hypothermia can lead to Na^+/K^+ ATPase alterations, ATP depletion, dysregulation of Ca^{2+} homeostasis, mitochondrial perturbations, xanthine oxidase accumulation, and increased levels of reactive oxygen species (ROS) which may have deleterious effects on cellular viability (2). Therefore, preservation solutions have been implemented in conjunction with hypothermia for additional cellular protection. Numerous solutions are commercially available while others remain institutionally derived.

There is continued uncertainty among clinicians regarding the most optimal preservation solution as evidenced by Demmy *et al.* who revealed the use of 167 different

solutions among United Network for Organ Sharing (UNOS) cardiac transplant centers (3). It is clear that investigation concerning the optimal preservation solution is necessary to reduce such widespread variability and potentially improve graft outcomes. As such, we sought to review the pertinent clinical studies available in an attempt to identify characteristics of an ideal preservation solution for both cardiac and pulmonary grafts with the intention of ultimately minimizing graft dysfunction and improving patient outcomes.

Classification of preservation solutions

Preservation solutions

Euro Collins (EC) solution was designed in the 1960s and considered the preservation solution of choice for over 15 years until organ perseveration was revolutionized by the introduction of University of Wisconsin (UW) solution in 1988 (4). However, the high molecular weight compounds within UW such as hydroxyethyl starch (HES) resulted in a highly viscous solution that was implicated in part, to organ dysfunction thereby, supporting the development of less vicious alternatives including Celsior (CEL) and histidine-tryptophan-ketoglutarate (HTK) (5).

Many targeted approaches to cardiac organ preservation have been attempted including Plegisol which arose from the initial St. Thomas solution used for cardioplegia, albeit with slight modifications including the addition of a buffering system (6). In contrast to the aforementioned acellular approaches, Papworth solution was centered on the inclusion of donor blood in its composition (7). The different metabolic demand and physiology of the lung supported the construction of pulmonary specific solutions including Perfadex (PER) which still remains confined for sole use in pulmonary transplantations by the Federal Drug Administration (FDA) in the United States.

Preservation solutions are composed of multiple elements, each with their own advantages and disadvantages (*Table 1*). We will highlight a common classification scheme for EC, UW, HTK, CEL, PER, Papworth, and Plegisol according to respective molecular properties.

Intracellular/extracellular

Preservation solutions can be broadly classified into intracellular and extracellular solutions based upon the potassium and sodium concentrations. Intracellular solutions closely recapitulate the high potassium/low sodium conditions present within the cellular milieu to minimize potential concentration gradients across the plasma membrane that could favor potassium efflux. UW and EC are popular intracellular solutions, however the perceived risk of hyperkalemia induced pulmonary vasoconstriction favored the design of extracellular (low potassium) solutions such as HTK, CEL, PER, Papworth, and Plegisol (10). Over time, intracellular and extracellular solutions were shown to be equivalent (10).

Impermeant/colloid

Hypothermia causes dysregulation of the Na^+/K^+ pumps in the cellular membrane resulting in cellular edema through sodium and water influx into the cell (12). The addition of an impermeant or colloid creates an osmotic force that preferentially promotes water retention in the extracellular compartment to counteract this effect. EC contains a high concentration of glucose that was intended to act as impermeable barrier. However, glucose is suboptimal as enzymatic cleavage occurs resulting in substrate diffusion into the cell and subsequent cellular edema (2). The development of newer solutions containing alternate impermeants/colloids led to superior protection against cellular swelling. UW contains lactobionate and the trisaccharide impermeant raffinose as well as the synthetic colloid HES (Roskott *et al.*). HTK, CEL, and Papworth rely on mannitol to combat tissue edema (9). In addition to mannitol, lactobionate and albumin are included in CEL and Papworth, respectively for further protection (9,11).

Buffer

Many of the commercial preservation solutions contain a buffer to combat the effects of metabolic acidosis that result from the shift of aerobic to anaerobic metabolism during periods of ischemia. UW, PER, and EC utilize phosphate buffers whereas, HTK and CEL are comprised of histidine buffering systems to prevent cellular damage (8,9). Bicarbonate is an effective buffer and used in EC and Plegisol (6,8).

Antioxidants

ROS are an inevitable consequence of tissue ischemia during the *ex vivo* period and can lead to significant cellular damage. UW counteracts ROS with a combination of allopurinol to inhibit the formation xanthine oxidase and glutathione which can act as a reducing agent (9). Glutathione is also the mainstay of antioxidant activity in CEL (9). HTK's antioxidant properties are attributed to tryptophan which is a functional electron donor (10).

Table 1 Comparison of select perfusate solutions

	EC	UW	HTK	CEL	PER	Papworth	Plegisol
Study	Aziz (8)	Roskott (9)	Roskott (9), 't Hart (10)	Roskott (9), 't Hart (10)	Aziz (8)	Marasco (11), Divisi (7)	Chambers (6)
IC/EX	IC	IC	EX	EX	EX	EX	EX
Na^+	10	25	15	100	138	115	120
K^+	115	120	10	15	6	3	16
Impermeant/ colloid	Glucose	LactoB, raffinose, HES	Mannitol	LactoB, mannitol	Dextran	Mannitol, albumin	–
Buffer	Phos, bicarb	Phos	Histidine	Histidine	Phos	–	Bicarb
Antioxidant	–	AlloP, GSH	Trp, mannitol	GSH, mannitol	–	Mannitol	–
Osmolarity (mOsm/L)	375	330	310	320	292	440	320
Ca^{2+}	–	–	0.02	0.25		Und	1.2
Mg^{2+}	–	5	4	13	0.8	–	16
Cl^-	15	20	32	–	142	Und	160
Glucose	180	–	–	–	5	–	–
Others			α-KG		SO_4^{2-} 0.8, dextran 40 g/L	Donor blood heparin	–

All units expressed in mmol/L unless otherwise indicated. Abbreviations: IC, intracellular; EX, extracellular; EC, Euro Collins; UW, University of Wisconsin; HTK, histidine-tryptophan-ketoglutarate; CEL, Celsior; PER, Perfadex; Und, undetermined; LactoB, lactobionate; HES, hydroxyethyl starch; Phos, phosphate; Bicarb, bicarbonate; GSH, glutathione; AlloP, allopurinol; Trp, tryptophan; α-KG, ketoglutarate.

Moreover, mannitol has been suggested to have antioxidant properties which may confer a benefit to CEL, HTK, and Papworth (10).

Heart transplantation

Although ischemia times as long as 13 hours have been reported for heart transplants, cold ischemia times are usually limited to less than 6 hours (13,14). CEL was initially a favorable extracellular preservation solution for heart transplants with several studies supporting its use (*Table 2*). A prospective study containing 70 patients revealed a safe role for CEL as a preservation solution in the setting of heart transplants with a 30-day survival of 91.4% and acute graft failure rate of 10% (15). This was supported by De Santo *et al*. who found an in-hospital mortality rate of 8% and 1 year mortality rate of 12% in 200 patients that received CEL (16). Interestingly, upon stratification into low and high risk grafts in that study, there was no difference in mortality or graft failure suggesting a potential safe role for the use of CEL even in the setting of prolonged ischemia (>180 minutes) (16).

Given the suggested beneficial role of CEL, many comparison trials were performed. An evaluation of 48 patients (24 HTK and CEL 24) suggested a beneficial role for CEL as only one case of graft failure was observed in the CEL arm compared to two in the HTK group. However, the results of this study were preliminary and the low number of patients made it difficult to derive any meaningful conclusions (17). Vega *et al*. (18) evaluated 131 patients with the use of CEL (n=64) to several other solutions (n=67) including: UW, Plegisol, Stanford solution, PlasmaLyte A, Carmichael solution, Roe, lactate ringers, and normal saline. There was no difference in the mortality rate at 30 days (CEL 94% *vs*. others 88%) or graft failure rate at 30 days (CEL 6.3, Cntrl 13.4%; P not listed) (18). Although comparisons of CEL to the use of a specific solution could not be made given the variety of controls in this study, it did once again demonstrate a safe use for CEL in heart transplants. To compare CEL against a limited number of control preservation solutions, Cannata *et al*. (19) evaluated 133 patients (CEL 38, HTK 61, and Plegisol 34) and found

Table 2 Selected clinical studies involving cardiac perfusate solutions

Study	Solution	Cases	Patient survival	Graft failure
Remadi (15)	CEL	70	91.4% (30 d)	10%
De Santo (16)	CEL	200	88% (1 y)	–
Wieselthaler (17)	CEL *vs*. HTK	48 (CEL 24, HTK 24)	No diff (CEL 4.2%, HTK 8.3%; P not listed)	No diff (CEL 4.2%, HTK 8.3%)
Vega (18)	CEL *vs*. several	131 (CEL 64, Cntrl 67)	No diff (30 d) (CEL 94%, Cntrl 88%; P not listed)	No diff (30 d) (CEL 6.3%, Cntrl 13.4%; P not listed)
Cannata (19)	CEL *vs*. HTK *vs*. Pleg	133 (CEL 38, HTK 61, Pleg 34)	No diff (in-hosp) (CEL 89.5%, HTK 83.7%, Pleg 85.3%; P=0.717)	No diff (CEL 10.5%, HTK 14.7%, Pleg 14.7%; P=0.814)
Kofler (20)	UW *vs*. HTK	340 (UW 118, HTK 222)	UW > HTK (UW 80.1%, HTK 66.1%; P<0.001)	–
George (21)	UW *vs*. CEL	174 (UW 42, CEL 132)	No diff (1 y) (UW 79.5%, CEL 80.3%; P=0.92)	UW > CEL (UW 0.0%, CEL 10.6%; P=0.02)
Garlicki (22)	UW *vs*. CEL *vs*. HTK	224 (UW 64, CEL 28, HTK 132)	No diff (90 d) (UW 84%, CEL 86%, HTK 88%; P not listed)	UW 9.4%, CEL 0.0%, HTK 4.5%; P not listed
George (23)	UW *vs*. CEL	4,910 (UW 3,107, CEL 1,803)	UW > CEL (UW 89.6%, CEL 87%, P<0.01)	–

Abbreviations: Cntrl, control; no diff, no statistically significant difference; UW, University of Wisconsin; CEL, Celsior; HTK, histidine-tryptophan-ketoglutarate; Pleg, Plegisol; in hosp, in-hospital.

no statistical difference with respect to in-hospital mortality [CEL 10.5%, HTK 16.3%, and Plegisol (St. Thomas) 14.7%, P=0.717] or graft failure (HTK 14.7%, CEL 10.5%, and Plegisol 14.7%, P=0.814).

UW emerged as a popular alternative for heart transplants as there was a survival benefit associated with its use compared to other solutions such as HTK. Kofler *et al.* (20) saw an improvement in survival after switching from HTK to the use of UW in their heart transplant series (UW 80.1% *vs.* HTK 66.1% survival at 1 year, P<0.001). During the transition to UW, that institution also began using nitric oxide and prostanoids to prevent right heart failure which may have imposed confounding effects. An evaluation of 174 patients (42 UW and 132 CEL) found no difference in 30-day/1 year mortality and primary graft dysfunction (UW 11.9% *vs.* CEL 26.5% P=0.059) with the use of UW (21). However, a higher rate of right heart failure was found in the CEL group (UW 0% *vs.* CEL 10.6% P=0.02) (21). Conflicting results were found in an evaluation of 224 patients (UW 64, HTK 132, and CEL 28) where a trend towards lower mortality at 90 days with the use of HTK was observed (UW 16%, HTK 12%, and CEL 14%) (22). Acute graft failure did not occur in the CEL group and was moderate in the UW and HTK groups (UW 9.4%, HTK 4.5%, CEL 0%; P not listed) (22).

The largest study to date was performed by George *et al.* (23) which addressed the mixed results observed between UW and CEL. It comprised 4,910 patients (UW 3,107 and CEL 1,803) and revealed an improvement in 1 year survival with the use of UW (UW 89.6% *vs.* CEL 87.0% P<0.01) (23). Graft survival was not stated (23). Although the improvement in survival is modest, it may account for the lack of statistically significant differences observed by George *et al.* (21) and Garlicki *et al.* (22) as these studies had relatively lower numbers of patients. Together these results suggest that UW should be the preservation solution of choice in heart transplants.

Lung transplantation

The lung can only tolerate a short period of ischemia, usually less than 6 hours (24). Tierney *et al.* (12) reported their experience with lung transplants over a one year duration using EC and prostaglandin E1 with a one year survival of 79%. Oto *et al.* (25) showed no difference in 30-day mortality in 157 lung transplants with the use of EC, Papworth, or PER. However, a follow up study at the same institution with a greater number of patients showed an

Table 3 Selected clinical studies involving lung perfusate solutions. Euro-Collins *vs.* Perfadex/low potassium dextran solutions

Study	Solution	Cases	Patient survival	PaO2/FiO2	Wean from ventilator
Aziz (8)	EC *vs*. PER	69 (EC 37, PER 32)	No diff (30 d) (EC 89.2%, PER 90.7%; P=0.88)	No diff (EC 244 mmHg, PER 266 mmHg; P=0.9)	No diff (EC 71.2 hr, PER 81.9 hr; P=0.4)
Gámez (31)	EC *vs*. PER	136 (EC 68, PER 68)	No diff (30 d) (EC 78, PER 80; P not listed)	No diff (EC 238 mmHg, PER 257 mmHg; P not listed)	No diff (EC 182 hr, PER 174 hr; P not listed)
Müller (32)	EC *vs*. PER	80 (EC 48, 32 PER)	No diff (30 d) (EC 88%, PER 94%; P not listed)	–	No diff (EC 3 d, PER 4 d; P=0.67)
Rabanal (33)	EC *vs*. PER	46 (EC 21, PER 25)	No diff (30 d) (EC 88%, PER 100%; P not stated)	PER > EC (PER 310 mmHg, EC 170 mmHg; P<0.05)	PER > EC (PER 72 hr, EC 92 hr; P<0.05)
Strüber (34)	EC *vs*. LPD	106 (EC 63, LPD 57)	No diff (EC 86%, LPD 92%; P not listed)	No diff (EC 282 mmHg, LPD 303 mmHg; P not listed)	PER > EC (EC 321 hr, LPD 189 hr; P=0.006)
Fischer (35)	(EC *vs*. PER) + PGE1	94 (EC 46, PER 48)	No diff (EC 89.6%, PER 93.5%; P=0.082)	PER > EC (EC 310 mmHg, LPD 370 mmHg; P=0.017)	–

Abbreviations: no diff, no statistically significant difference; PGE1, prostaglandin E1; EC, Euro-Collins; PER, Perfadex; LPD, low potassium dextran; d, day; hr, hours.

increased correlation with long-term death associated with the use Papworth compared to EC or PER in 310 patients [216 double lung transplantations (DLT) and 94 single lung transplantations (SLT)] (11). The effect on mortality is not apparent until after 3 years, potentially accounting for the lack of difference observed among the three perfusate solutions in the Oto's study (25). In both studies there was a lower incidence of primary graft dysfunction observed with PER (11,25). In a larger study comparing multiple solutions, Ganesh *et al.* (26) found no difference in risk adjusted mortality among 681 patients who received EC (284 patients), blood albumin [139], low potassium dextran (LPD) solution (commercially sold as PER), or core cooling (107 patients).

Intracellular preservation solutions were initially used in lung transplants. Hardesty *et al.* (27) compared the use of EC (30 patients) to UW (70 patients) in 100 transplants [13 heart-lung (HLT), 45 DLT, 42 SLT transplants). Both solutions were found to be comparable (27). Given the potential for pulmonary dysfunction from potassium induced vasoconstriction with intra-cellular solutions, extracellular preservation solutions became a topic of interest (28). Thabut *et al.* (29) evaluated 170 patients (124 SLT and 46 DLT) who received UW, EC, Cambridge, or CEL (n=24, 61, 64, and 21 patients, respectively). There was no difference in 1 month mortality however, there was a lower incidence of post-transplant graft edema with the use of Cambridge solution (an extracellular solution) after adjustment for the duration of graft ischemia (29). One of the largest comparison studies involving the use of UW in lung transplants was performed by Arnaoutakis *et al.* (30) who evaluated 4,455 patients (4,161 LPD *vs.* 294 UW) and found an increased risk of mortality at one year with the use of UW (hazard ratio 1.75, P=0.004) after multivariate analysis.

EC has been directly compared to PER (a LPD) in multiple studies (*Table 3*). Aziz *et al.* (8) compared the use of EC and PER in 69 patients (EC 37 and PER 32). There were 12 SLT (EC 7, PER 5), 51 DLT (EC 27, PER 24), and 6 HLT (3 EC, PER 3) (8). There was no difference in the 30-day mortality (EC 10.8% *vs.* PER 9.3%, P=0.88), PaO2/FiO2 ratio (EC 244 *vs.* PER 266 mmHg, P=0.9), or duration of mechanical ventilation (EC 71.2 *vs.* PER 91.9 hr, P=0.4) (8). Similar results were observed by Gámez *et al.* (31) who compared the use of EC to PER in 136 lung transplants [SLT (EC 32, PER 15) and DLT (EC 36, PER 53)] and found no difference in 30-day mortality, length of time on the mechanical ventilator, and PaO2/FiO2 ratio (P values not listed). However, the EC group had a higher incidence (EC 37% *vs.* PER 16%, P=0.01) of severe graft failure (PaO2/FiO2 <150 mmHg) despite a higher number of double lung transplant recipients in the PER group (31).

These results have been refuted by several other studies that have suggested differences between EC and PER. Müller *et al.* (32) evaluated 80 patients who received either EC or PER [46 SLT (EC 31 and PER 15) and 34 DLT (EC 17 and PER 17)]. There was a trend towards improved 30-day mortality (EC 12% *vs.* PER 6%, P not listed) and 1 year

mortality (EC 62% *vs.* PER 79%, P not listed) associated with the use of PER (32). PER was also associated with a favorable reperfusion injury score and improved alveolar/arterial oxygen ratio while the duration of mechanical ventilation was not statistically significant (P=0.67) (32). Rabanal *et al.* (33) evaluated 46 patients undergoing lung transplantation who received EC or PER (EC 21, PER 25 patients). There was no statistical difference in the 30 day mortality between both groups (EC 12% and 0% PER, P not stated), however, there was a better PaO2/FiO2 ratio (EC 170 *vs.* PER 310, P<0.05) and lower duration of mechanical ventilation (EC 92 EC *vs.* PER 72, P<0.05) associated with the use of PER (33). In similar comparisons, Fischer *et al.* (35) also observed a lower PaO2/FiO2 (EC 310, LPD 370 mmHg; P=0.017) with the use of PER while Strüber *et al.* (34) observed a shorter duration of mechanical ventilation (EC 321 *vs.* LPD 189 hr, P=0.006) that correlated with the use of a LPD solution such as PER. Of note, the duration of mechanical ventilation in the Strüber (34) study was substantially longer than other studies such as Rabanal *et al.* (33).

Together these studies suggest against the use of Papworth and UW as they may impose an increased risk of mortality. In comparing two of the most commonly used extracellular preservation solutions in lung transplantation (EC and PER) there does not appear a survival benefit afforded with the use of either solution. However, the improved PaO2/FiO2 and lower duration of mechanical ventilation observed in some studies favor the use of PER.

Conclusions

Based upon the aforementioned studies, UW is superior for cardiac transplantation with a slight survival advantage compared to CEL while PER is the preferred solution for pulmonary transplantations. The use of PER correlates with an improved PaO2/FiO2 ratio and a shorter duration of mechanical ventilation. While we looked at graft survival and overall patient survival, it should be noted that these outcomes are not solely dependent on the preservation solution used. Several variables such as the quality of the graft, surgical technique, and immunosuppression regimen have important contributions to the overall success. Additionally, the survival time point used in our review may not have encompassed the long-term effects associated with the use of a particular preservation solution. Many of the studies were also limited by small sample sizes and may have been underpowered to detect minute differences. The optimal preservation solution for each respective organ can be supported by available evidence based data and might be a useful adjunct to ameliorate the widespread viability observed by Demmy *et al.* (3) among different centers.

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