

## ORIGINAL ARTICLE

# A single solution for multiple organ procurement and preservation

Georges Karam,<sup>1</sup> Philippe Compagnon,<sup>2</sup> Maryvonne Hourmant,<sup>1</sup> Philippe Despins,<sup>3</sup> Daniel Duveau,<sup>3</sup> Didier Noury<sup>4</sup> and Karim Boudjema<sup>2</sup>

1 Pôle Néphrologie-Urologie-Transplantation, Hôtel Dieu, Centre Hospitalier Universitaire de Nantes, Nantes, France

2 Département de Chirurgie Viscérale, Hôpital Pontchaillou, Centre Hospitalier Universitaire de Rennes, Rennes, France

3 Pôle Thorax, Hôpital Nord, Centre Hospitalier Universitaire de Nantes, Nantes, France

4 Etablissement Français des Greffes, Service de Régulation et d'Appui Région Ouest, Hôpital Pontchaillou, Rennes, France

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## Correspondence

Karim Boudjema MD, PhD, Département de Chirurgie Viscérale, Hôpital Pontchaillou, Centre Hospitalier Universitaire de Rennes, 35033 Rennes, France. Tel.: +33-299-28-41-01; fax: +33-299-28-41-29; e-mail: karim.boudjema@chu-rennes.fr

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## Summary

Two or three different solutions may be used to preserve thoracic and abdominal organs during a single procurement. The aim of this prospective, multi-center, noncomparative study was to evaluate the safety and efficacy of Celsior® (study solution, solution S) as a flushing and cold storage solution for both thoracic and abdominal organs. Between August 1999 and July 2000, 72 consecutive multiple-organ procurements were performed using solution S as the sole solution for flushing out and cold-storing thoracic and abdominal grafts. Two hundred and sixty-four grafts were implanted into 245 recipients (131 kidneys, 9 kidney–pancreases, 69 livers, 34 hearts and 6 heart–lungs). The mean cold ischemia time was 21 h for kidneys (26% > 24 h); 11 h 26 min for pancreases, 9 h 16 min for livers (23% > 12 h), and 2 h 58 min for hearts and lungs. No cardiac failure or arrhythmia occurred on graft reperfusion. Fourteen percent of kidney recipients had delayed graft function. The mean serum creatinine level at 3 months was  $123 \pm 41 \mu\text{mol/l}$ . All pancreas recipients were insulin-free at 3 months. Primary graft nonfunction occurred in one liver recipient. Complete hepatic artery thrombosis occurred in six liver recipients during the first month; four of these patients had a risk factor for thrombosis. All but three of the heart recipients were in sinus rhythm on day 1, and 65% were extubated on day 1. Inotropic drugs were necessary during the first 72 h in 25% of heart recipients. Twelve-month patient and graft survival rates were, respectively, 100% and 96% (kidney), 100% and 89% (pancreas), 88% and 83% (liver), 77.5% (heart) and 67% (heart–lung). These results suggest that Celsior®, a ready-to-use solution, is safe and effective for multiple organ procurement and preservation.

## Introduction

Graft viability during ischemic transfer from donor to recipient is mainly based on hypothermia, which is initially achieved by flushing the organs with cold (4 °C) preservation solution while still *in situ*. Preservation solutions are formulated to attenuate the effects of ischemia, and their efficacy depends on both their composition and

the type of organ. The quality of organ preservation is a major determinant of initial graft function and survival.

Belzer UW cold storage solution (Viaspan®; Du Pont Pharmaceuticals, DE, USA) is the reference solution for preserving abdominal organs, i.e. liver [1,2] kidney [3] and pancreas [4]. It is an 'intracellular' solution containing a high potassium concentration (130 mmol/l). Its efficacy is largely due to the presence of high-molecular-

weight impermeants (lactobionic acid and raffinose, which inhibit intracellular oedema secondary to ischemia), and agents (glutathione and allopurinol), which attenuate the deleterious effects of free radicals, which are produced in large amounts on reperfusion [5]. UW solution is less frequently used for preserving thoracic organs [6]. Hearts can be preserved with St Thomas's solution [7], HTK [8] or Euro Collins solution [9], and lungs with Collins solution or Papworth solution, a homemade preparation containing donor blood [10]. Thus, two or even three different solutions may be used to preserve thoracic and abdominal organs during a single procurement, increasing both the complexity and the cost of the procedure.

Celsior® (Imtix-SangStat, Lyon, France; referred to below as solution S), an 'extracellular' solution containing a high sodium concentration (100 mmol/l), is effective for cold preservation of hearts [11] and lungs [12]. Like UW solution, solution S contains high-molecular-weight impermeants (lactobionic acid and mannitol), free-radical scavengers, and reduced glutathione. Histidine is added to buffer intracellular acidosis [13], and contributes to limiting calcium overload [14]. The compositions of UW and solution S are compared in Table 1. In laboratory experiments, Celsior has proved suitable for preserving liver [15], kidney [16], and pancreas [17].

The aim of this prospective clinical study was to assess the safety and efficacy of Celsior® when used as the sole

solution for flushing and cold-storing both thoracic and abdominal organs.

## Patients and methods

Between August 1999 and August 2000, all organ procurements carried out in the Ouest region of France (Region 6 of the French organ-sharing organization) by teams at the university hospitals of Rennes, Nantes, Brest, Limoges, Poitiers and Caen were performed using only solution S to flush and cold-store both thoracic (heart and lungs) and abdominal organs (liver, kidney and pancreas).

## Procurement and cold storage

The characteristics of the donors and the type and number of organs procured are shown in Table 2. Procurement was carried out using a standard technique for both abdominal and thoracic organs, as originally described by Starzl *et al.* [18]. Abdominal organs were flushed with 4–6 l of solution S via the aorta. Livers were flushed with an additional 2 l via the inferior mesenteric vein, and the biliary tract was rinsed *ex situ* with a further 100 ml. Thoracic organs (heart and lungs) were harvested first. Livers and pancreases were harvested 'en bloc' next, and separated *ex vivo*. This allowed a shortening of the kidney warm ischemia time, these were harvested separately and

**Table 1.** Comparative formulation of Celsior® (study solution) and Viaspan® (UW solution).

Main components	Celsior®	Viaspan®
Electrolytes (mmol/l)		
Sodium	100	30
Potassium	15	130
Magnesium	13	5
Calcium	0.25	–
Impermeants (mmol/l)		
Mannitol	60	–
Lactobionic acid	80	100
Raffinose	–	30
Oncotic agents (g/l)		
Hydroxyethyl starch	–	50
Antioxidants (mmol/l)		
Glutathione	3 (reduced)	3 (total)
Allopurinol	–	1
Energy precursors (mmol/l)		
Glutamic acid	20	–
Adenosine	–	5
Buffers (mmol/l)		
Histidine	30	–
Phosphate	–	25
Osmolality (mOsmol/l)	320	320
pH	7.3 ± 0.1	7.4 ± 0.1

**Table 2.** Donor characteristics and organ procurement.

Donor characteristics	
Age (years; mean ± SD; range)	33.8 ± 13.4 (14–71)
Sex ratio M/F	48/24
Causes of death (%)	Trauma 51, vascular 39, other 10
ICU stay (h; mean ± SD; range)	64 ± 55 (10–264)
Reversible cardiac arrest (%)	26
Use of inotropic agents (%)	71
Anuria >3 h (%)	4
Procurement characteristics	
<i>n</i> (%)	
Organ combinations removed	
Kidneys/liver/heart	28 (39)
Kidneys/liver	27 (38)
Kidneys/liver/heart/lungs	5 (6.5)
Kidneys/liver/pancreas	5 (6.5)
Kidneys/liver/pancreas/heart	3 (4)
Kidneys/heart	2 (3)
Kidneys/liver/pancreas/heart/lungs	1 (1.5)
Liver/heart	1 (1.5)
Total number of donors	72
Organs removed	
Kidney	140
Liver	69
Heart	40
Pancreas	9
Lung	6
Total number of organs	264

last. The inferior vena cava was left entire together with the right kidney. An aortic patch was harvested each kidney. All organs were placed in solution S at 4 °C until transplantation.

### Transplantation

The six *en bloc* heart–lung grafts and the 34 heart grafts were implanted as quickly as possible to minimize the cold ischemia time. The nine whole-pancreas grafts were implanted intraperitoneally with enteric and venous systemic drainage, simultaneously with a kidney. The 69 whole-liver grafts were implanted orthotopically, either immediately ( $n = 26$ ) or the morning after if harvested at night ( $n = 43$ ). One hundred and thirty-one kidneys were transplanted electively, following negative cross-matching.

The general characteristics of the recipients are shown in Table 3. All organs were implanted with the standard surgical techniques used in each center. Liver grafts were flushed with 500 ml of cold (4 °C) 4% human albumin via the portal vein immediately before revascularization.

The participating centers received no special instructions regarding immunosuppressive regimens for the purposes of this study.

### Evaluation criteria

The efficacy of the solution was judged on the following criteria: the incidence of primary graft nonfunction; the time to normal graft function; the frequency of immediate vascular complications (venous or arterial thrombosis); and the incidence of biliary complications in liver recipients. Graft function was judged as follows: *heart and lung grafts*: need for inotropic agents, duration of mechanical ventilation, and changes in blood gas levels ( $\text{PaO}_2$  and  $\text{PaCO}_2$ ); *kidney grafts*: incidence of postoperative dialysis and changes in serum creatinin levels; *liver grafts*: bile production, transaminase levels, bilirubin levels, and prothrombin time; *pancreas grafts*: insulin requirements and C peptide levels. Patient and graft survival rates were calculated at 12 months.

The safety of solution S was evaluated on the basis of the incidence of cardiac arrhythmias or cardiac arrest

after graft reperfusion; the incidence of aerobic and anaerobic bacterial growth in preservation solution sampled at the end of the cold storage period; and the incidence of infections due to the same microorganisms in the graft recipient.

### Statistical analysis

Data were entered into a centralized database using SPSS software (SPSS Inc., Chicago II, USA). All patients were followed until death or retransplantation, or for at least 1 year after transplantation. An interim analysis was carried out at 3 months. Qualitative data are expressed as percentages. Quantitative data are expressed as mean  $\pm$  SEM. Graft and patient survival rates were calculated using the Kaplan–Meier method.

## Results

### Heart and heart–lung grafts

The mean cold ischemia time (CIT) for the 34 hearts and six heart–lung grafts was 2 h 58 min  $\pm$  11 min (median 2 h 57 min, range 51 min–4 h 50 min). Samples of preservation solution taken prior to graft implantation grew no bacteria. Exactly 92.5% of transplanted hearts immediately entered sinus rhythm, and the systolic ejection fraction on day 1 exceeded 60% in all but one of the recipients. Sixty-five percent of patients were extubated within 24 h after grafting. On day 3, 75% of patients were free of inotropic support.

Forty-eight percent of the heart recipients developed bronchopulmonary infections, and 18% had at least one hemodialysis session for renal impairment. The  $\text{PaO}_2/\text{FiO}_2$  ratio was  $41.82 \pm 13.5$  on day 7. Seven (20.5%) of the 34 heart recipients and two (33%) of the six heart/lung recipients died during the first year. Six patients developed grade IB rejection and three patients developed grade II rejection between months 1 and 3. The 1-year patient survival rate was 77.5%.

### Liver grafts

Bacteria were cultured from 19% of preservation solutions (13/69). The same organism was isolated in two recipients, from bile in one case and the peritoneal cavity in the other case. The mean CIT was 9 h 16 min (6 h 44 min for livers implanted straight away, and 10 h 49 min for livers implanted electively the morning after harvesting). Sixteen grafts had a CIT of more than 12 h. No cardiac arrhythmias occurred during graft reperfusion.

Eight hepatic artery thromboses occurred in seven patients. In one case the thrombosis was limited to the

**Table 3.** Characteristics of the recipients.

	Kidney (pancreas)	Liver	Heart (heart/lung)
<i>n</i>	140 (9)	67	40 (6)
Mean age (years)	42 $\pm$ 13	49 $\pm$ 11	45 $\pm$ 14
Sex ratio M/F	1.75	2.2	4.7
First transplant (%)	83	94	100

left branch of the hepatic artery, and had no apparent effect on graft function or recipient survival. Thrombosis occurred in two consecutive grafts received by one patient. The mean CIT of these eight grafts was 9 h 20 min. A risk factor for hepatic artery thrombosis was found in five of the eight cases (Table 4). Arterial thrombosis caused six graft losses, resulting in the death of three patients.

One case of primary nonfunction occurred (1.5%). The donor had been in intensive care unit for 11 days and the CIT was 13 h 11 min. Sixty-one grafts functioned immediately; liver biochemistry values returned to normal from day 7 onwards and remained normal at month 1 and month 3. A biopsy-proven reversible episode of acute rejection was observed in 35% of cases. Delayed graft function occurred in seven cases (10%). The overall 1-year graft and patient survival rates were 83% and 88%, respectively.

### Kidney grafts

Bacteria were cultured from 21% of preservation solutions (29/140). The same organism was cultured from the urine of one recipient. The mean CIT was 21  $\pm$  8 h (median

19 h, range 4–45 h). No cardiovascular or hemodynamics events occurred during reinfusion. Diuresis exceeded 500 ml/24 h in 96% of patients on day 3. Delayed graft function occurred in 19 patients (14%), necessitating dialysis. The average serum creatinin levels were 225  $\pm$  222, 130  $\pm$  50 and 123  $\pm$  41  $\mu$ mol/l on day 7 and at months 1 and 3, respectively. Nine patients (6%) developed urinary tract infections. Five grafts were lost, owing to venous thrombosis in one case (0.7%) and to hyper acute vascular rejection in one case (a second transplant). The causes of graft loss are summarized in Table 5. Patient and graft survival rates at 1 year were 100% and 96%, respectively.

### Pancreas grafts

The mean CIT was 11 h 26 min  $\pm$  52 min (median 12 h 17 min; range 6–14 h 57 min). Bacteria were cultured from 19% of preservation solutions (2/9) but none of the recipients developed infections due to the corresponding organism. One pancreas was removed on day 2 after grafting because of venous thrombosis. Only one of the other eight recipients still needed insulin on day 3, and all the patients were insulin-free after 1 month. Two patients developed acute oedematous pancreatitis compli-

**Table 4.** Characteristics of liver grafts and recipients with hepatic artery thrombosis.

Case no.	Recipient age (years)	CIT	Donor age (years)	Risk factor for thrombosis
1	55	10 h 20 min	47	Recipient portal vein thrombosis and hepatic artery stenosis; double hepatic artery and portal vein jump PTFE vascular graft prosthesis.
2	55	12 h 40 min	46	Extensive arteriosclerosis, graft hepatic artery implanted on recipient atheromatous splenic artery.
3*	55	9 h	20	Retransplantation for hepatic artery thrombosis. Hepatic artery was implanted on recipient iliac artery.
4	58	6 h 9 min	58	Massive graft steatosis (80%)
5	48	5 h 10	32	None
6	55	12 h 28 min	14	Large liver graft
7	36	9 h	19	None
8	45	10 h	65	None (thrombosis of the left branch of the hepatic artery)

CIT, Cold ischemia time.

\*Same recipient as no. 2.

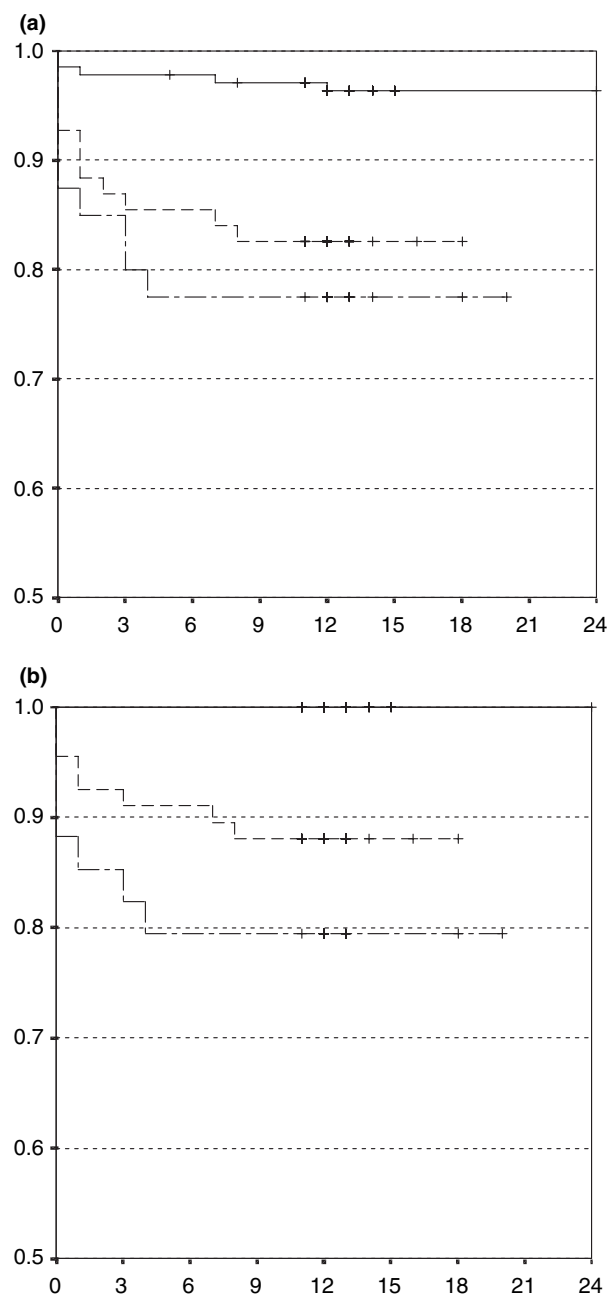
**Table 5.** Causes of kidney graft loss.

Case no.	Recipient age (years)	CIT	Donor age (years)	Cause of graft loss
1	43	28	28	Hyperacute vascular rejection, positive cross-match on historical sera
2	40	20 h 30 min	30	Venous thrombosis, no risk factor
3	14	12 h	46	Acute rejection
4	46	18 h	28	Chronic rejection
5	46	24 h 30 min	61	Acute pyelonephritis

CIT, Cold ischemia time.

cated by paralytic ileus. Both recovered after peritoneal lavage. At 3 months the mean fasting blood glucose and C peptide levels were  $4.6 \pm 0.9$  mmol/l and  $2.44 \pm 2.8$  ng/ml, respectively. The 1-year patient survival rate was 100%, and the kidney and pancreas graft survival rates were 100% and 89% (one graft loss), respectively.

Kaplan–Meier survival curves for the patients and the different graft types are shown in Fig. 1.



**Figure 1** Twenty four month graft (a) and patient (b) survival according to Kaplan–Meier. Kidneys (—), Livers (---) and hearts (···).

## Discussion

Thoracic and abdominal organs are preserved using a variety of solutions, according to the center and the specific organ. UW solution is commonly used for abdominal organs and rarely for thoracic organs. In contrast, solution S is used by many teams to preserve thoracic organs but rarely for abdominal organs, i.e. kidneys and livers. This pilot prospective but nonrandomized study was not designed to compare S solution to any other preservation solution. We used solution S alone to preserve both abdominal and thoracic organs, a strategy which has never been reported with any other preservation solution, and found it to be safe and effective.

Solution S was originally designed to preserve heart grafts, and its capacity to preserve lungs was recently demonstrated [19]. Our use of solution S to preserve abdominal organs was based on both theoretical and experimental considerations. First, the composition of solution S is very similar to that of other solutions used to preserve abdominal organs. It contains lactobionic acid and reduced glutathione, which are key components of UW solution [20], together with mannitol, an impermeant that scavenges free radicals [21], and histidine, a buffer essential for the performance of HTK solution [22]. Contrary to most organ-preservation solutions, solution S is an extracellular solution containing a high sodium concentration. High potassium concentrations have been shown to damage endothelial cells [23]. Moen *et al.* [24] showed that UW solution, in which the Na/K ratio is reversed, efficiently preserved canine abdominal organs (liver, kidney and pancreas). Preclinical studies have shown that solution S is also suitable for kidney, liver and pancreas preservation [15–17]. ‘Extracellular’ preservation solutions also contain high potassium concentrations (15 mmol/l in solution S), and may therefore carry a risk of provoking cardiac arrhythmia during graft reperfusion. No such effects occurred in our study, in which cardiac adverse events were a special focus of attention.

All current organ-preservation solutions are subject to microbial contamination and growth, the risk of which increases with the preservation time; abdominal organs are also associated with a higher risk than thoracic organs. In our study bacterial contamination of solution S only occurred with liver, kidney and pancreas grafts. A multimicrobial flora was found in almost all cases: *Staphylococcus epidermidis*, *Streptococcus* and gram-negative bacilli. This indicates contamination of the surgical field with microbes from the skin and bile/GI tract. Only three recipients (two liver and one kidney) became infected by the organism isolated from the corresponding preservation solution. There was no death or morbidity related to this contamination.

Graft quality was mainly assessed on the basis of patient and graft survival, which reflect both graft function and postoperative complications, including vascular thrombosis (possibly linked to endothelial damage caused by the preservation solution). The only case of primary nonfunction occurred in a liver recipient, who required retransplantation. The liver donor had spent a long time in intensive care, and the CIT was more than 12 h; both these factors have been linked to an increased risk of graft nonfunction [25,26].

Venous thrombosis was rare in this study, and caused the loss of only one renal graft and one pancreatic graft. Pancreas transplant patients are at risk of graft venous thrombosis associated with pancreatic necrosis [27], whatever the preservation solution. However, our subsequent experience since the end of this study did not show any higher risk of venous thrombosis and pancreatitis [28].

Graft venous thrombosis is a seldom event in renal transplantation [29], a setting in which increased donor age and CIT are two recognized risk factors. No clear cause of venous thrombosis was found in our patient whose graft was lost, as the CIT was short (20 h 50 min) and the donor was only 30 years old.

Complete thrombosis of the hepatic artery occurred in seven liver grafts (10%). This is a high rate compared to the 4–8% reported in other large series of adult liver transplantation [30,31]. However, two thromboses occurred in the same recipient, who may therefore have had a predisposing factor, and such factors were found in three of the other five patients. As hepatic artery thrombosis may be due to endothelial cell injury, a direct implication of the solution may not be excluded. We are now achieving a controlled study, specifically designed to evaluate the rate of hepatic artery thrombosis.

According to European registers, overall 12-month patient and graft survival rates are, respectively, 80.6% after heart transplantation (ISHLT Transplant Registry, 1997–2001), 94% and 82% after pancreas transplantation (International Pancreas Transplant Registry, 1997–2001), 79% and 72% after liver transplantation (European Liver Registry, 1988–2001), and 95% and 89.2% after kidney transplantation (OPTN data on January 1, 2002). The survival rates in our study are compatible with these European data.

In conclusion, this prospective, multicenter, noncomparative study suggests that Celsior® is a safe and effective solution for the procurement and preservation of both abdominal and thoracic organ grafts and enables a simplification and homogenization of procurement and preservation procedures. A prospective case-control study is ongoing to compare Celsior® and UW in liver preservation and to clarify the risk of arterial thrombosis observed.

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