

A Paired Study Comparing the Efficacy of Renal Preservation by Normothermic Autologous Blood Perfusion and Hypothermic Pulsatile Perfusion

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THE REDUCTION in metabolic rate effected by hypothermia constitutes the central tenet of traditional organ preservation.¹ The antithesis of this approach is to provide for metabolic demand during normothermic preservation. This latter strategy has two potential benefits. First, if metabolic requirements are met during preservation, then longer preservation with better posttransplant function may be achieved than for an organ suffering prolonged cold ischemia. Second, normothermic perfusion may allow *ex vivo* organ function to be restored and therefore assessed prior to transplantation. This may provide a basis for viability assessment, thus reducing the risks of primary nonfunction of allografts. POPS (pulsatile organ perfusion system) (Transmedics, Boston, Mass, USA) is a new warm perfusion system for organs, using blood as the principle component of the perfusate. The aim of this study was to compare the efficacy of preservation of porcine kidneys by normothermic autologous blood perfusion and hypothermic machine perfusion (RM3 system, Waters, Mass, USA.)

MATERIALS AND METHODS

Animals

Six large white pigs weighing 80 to 100 kg were sacrificed after electrical stunning followed by ex-sanguination, collecting the blood into 25,000 units of heparin. The kidneys were retrieved through an anterior abdominal approach; the retrieval commenced immediately when the cardiac output ceased. Once retrieved the kidneys were immediately perfused with cold (4°C) hyperosmolar citrate solution and stored on ice for the 2 hours necessary to transfer to the laboratory for perfusion.

Perfusion

One of each pair was perfused on the POPS system and the other on the Waters RM3 system as follows:

POPS

The POPS perfusion pressure was provided by an atraumatic pump, causing minimal erythrocyte lysis; the mean perfusate pressure was adjusted to 50 mm Hg. The perfusate was pumped through a heat exchanger, with the water bath set to maintain a perfusate temperature of 37°C, and a gas exchange unit, which maintained the pO₂ at 500 to 700 mm Hg. The gas for exchange was

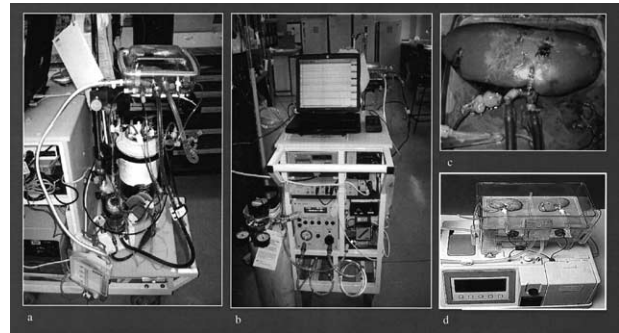


Fig 1. The perfusion apparatus systems. (a) The POPS circuit. (b) POPS control and monitoring and (c) the kidney in the organ chamber. (d) kidneys on the RM3 system.

96% O₂ and 4% CO₂, the latter for pH homeostasis. The perfusate was supplemented by infusions of nutrients and colloid to replace urinary losses. The warm oxygenated blood perfused the kidney through the cannulated renal artery and drained back into a venous reservoir through a cannulated renal vein. The ureter was cannulated and urine drained away from the circuit (see Fig 1a–c).

RM3

The RM3 system drives an acellular perfusate, Belzer's machine perfusion solution (MPS), with a mean pressure of 40 mm Hg, through a countercurrent cooling unit with the water reservoir kept cold with ice, maintaining a perfusate temperature of 3 to 8°C. The renal artery was cannulated for perfusion, with the renal vein effluent allowed to drain through the organ chamber back into a perfusate reservoir (see Fig 1d).

For both perfusion methods, the duration of perfusion was 16 hours, at the end of which their functions were assessed and compared. Cefuroxime (750 mg/L) was added to both perfusates to reduce the risk of microbial contamination.

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Table 1. Comparison Between the Kidneys Used in Both Groups

	Warm Ischemic Time (min)	Cold Ischemic time (hr)	Kidney Weight (g)
POPS	8 (6–10)	2.1 (1.7–2.3)	220 (190–300)
RM3	8 (6–10)	2.0 (1.7–2.2)	250 (210–310)

Assessment of Function

At the end of the preservation period the kidneys were removed from their machines and perfused with cold (4°C) hyperosmolar citrate solution. Then each was transferred to a POPS apparatus and perfused as above for 2 hours. During this assessment period, the ex vivo function of the kidneys was made by measuring urine output, perfusate flow rate and pressure, and urine and perfusate concentrations of sodium, creatinine, protein, and glucose. From these the intrarenal vascular resistance and urine-to-plasma concentration ratios of sodium and creatinine were calculated. The creatinine clearance was also calculated by multiplying the creatinine concentration ratio by the urine output (mL/min).

Statistics

Results are median (range) and statistical comparison was performed using the Mann-Whitney *U* test. Significance is taken at the $P = .05$ level.

RESULTS

The warm ischemic times (from death to organ flushing) and cold ischemic times and kidney weights for both groups are compared in Table 1. There are no significant differences between the groups.

The sodium and creatinine ratios, creatinine clearances, proteinuria, glycosuria, and intrarenal vascular resistances were compared between the groups. The ability of kidneys to concentrate creatinine and conserve sodium were significantly better in the POPS-preserved kidneys than in the RM3-preserved kidneys ($P = .03$ for both) (Fig 2a,c). Creatinine clearance was higher; vascular resistance, proteinuria, and glycosuria were lower in the POPS-preserved kidneys, although for these parameters the differences between the groups did not reach significance ($P = .7, .3, .3$, and $.2$, respectively) (Fig 2b to 2f).

DISCUSSION

The POPS-preserved kidneys performed better than the RM3-preserved organs, as measured by ex vivo indices of function. For creatinine and sodium concentration ratios these differences were significant.

The study used paired kidneys so that the groups were closely comparable. Larger groups may determine whether the other parameters measured were significantly different. Hypothermic pulsatile perfusion preservation is thought to be superior to static cold storage, particularly for kidneys retrieved from non-heart-beating donors.²

Brasile et al³ have used warm perfusion with an acellular perfusate containing perfluorochemical emulsion to in-

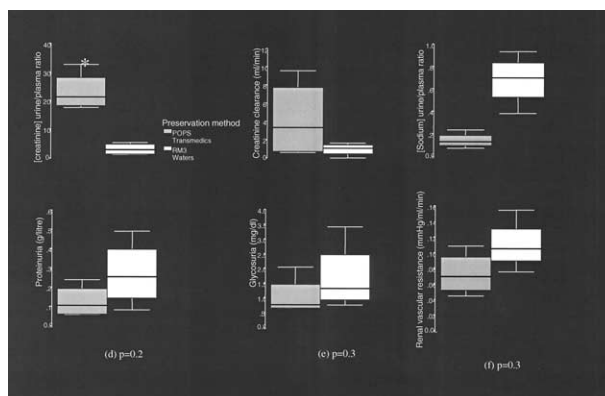


Fig 2. Results: (a) creatinine concentration ratios (b) creatinine clearances (c) sodium concentration ratios (d) glycosuria (e) proteinuria (f) intra-renal vascular resistance.

crease oxygen carriage for renal preservation. They report improved renal transplant function of ischemically damaged organs (simulating non-heart-beating donors) compared with conventional hypothermic perfusion.

Hassanein et al⁴ report encouraging results using the POPS for ex vivo preservation of porcine hearts and the ex vivo measurement of cardiac function.

Imber et al⁵ used autologous blood perfusion to preserve porcine livers, again with improved ex vivo measurements of function (bile secretion) compared with conventional hypothermic techniques.

Normothermic organ preservation may have the advantages of improving the preservation itself and allowing pretransplant evaluation of function. These attributes would be particularly useful for organs procured from non-heart-beating donors, as these already suffer inevitable warm ischemic damage prior to retrieval and so tolerate additional damage due to poor preservation.⁶ Also because of the warm ischemic insult they suffer, there is a pressing need for reliable objective preoperative viability testing.

While the results are very encouraging, transplant experiments are required to confirm that the ex vivo measurements of function correlate with posttransplant in vivo function. Autotransplant experiments similar to those described by Brasile would be ideal, as any alloimmune response could be eliminated as a cause for posttransplant organ dysfunction.

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