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ORIGINAL ARTICLE

The effect of inadequate *in situ* perfusion in the non heart-beating donor

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Summary

In situ aortic perfusion in the nonheart-beating donors (NHBD) is an important procedure to reduce primary warm ischaemic injury prior to formal donor organ retrieval. It allows an interim period to obtain donor family consent and theatre preparation. This study describes our experience of inadequate aortic perfusions resulting from difficult aortic cannulations and associated adverse outcome despite reasonable viability tests. Since 1998, all NHBD in our institution are perfused *in situ* using a double balloon triple lumen (DBTL) catheter inserted through a femoral artery cut-down procedure. The DBTL catheter is positioned with distal occlusive balloon at the aortic bifurcation using the 'pull-back' technique, the proximal occlusive balloon lies above the renal arteries. This provides selective aortic perfusion in particular the kidneys. Venous decompression using a femoral vein catheter enables a 'two-way infusion system'. Pre-transplant viability status of retrieved kidneys is determined by measuring pressure/resistance characteristics to the flow and biochemical markers for ischaemic injury. There were 90 NHBD renal transplants performed from 72 donors. Three renal transplants were carried out from three donors of ineffective *in situ* perfusion secondary to cannulation difficulties. Femoral cannulation was difficult as a result of extensive atherosclerosis of donor vessels. The comparison of allograft outcome from effective and ineffective *in situ* perfusion of donors showed high rate of primary nonfunction (PNF) from ineffective perfusion (chi-squared, $P < 0.0001$). The cases demonstrated poor outcome from ineffective perfusion related to the cannulation difficulties. Therefore a strict policy should be taken in cases where aortic cannulation and perfusion is inadequate, despite pretransplant assessment. In these circumstances, the primary warm ischaemia time should be extended to include this period of ineffective perfusion.

Introduction

The use of nonheart-beating donors (NHBD) to expand the kidney donor pool is becoming widely encouraged by transplant registries [1]. NHBD are associated with warm ischaemic injury at the time of cardiac arrest. Primary warm ischaemia is the period from cardiac arrest until intravascular cooling by either *in situ* or direct perfusion of the allograft. Preservation techniques used for NHBD kidneys are based on cooling manoeuvres such as intravascular cooling [2], intraperitoneal cooling [3], total body cooling and hypothermic machine preservation [4].

Since the development of the double balloon triple lumen (DBTL) catheter in 1975, selective aortic perfusion has been possible in the haemodynamically unstable donor [5]. This has permitted renal wash-out and cooling to temperatures below 18 °C [6], enabling significant reduction in renal metabolic demands [5]. *In situ* aortic

perfusion in the NHBD is essential to bridge the gap between cardiac arrest and donor nephrectomy. It provides the time needed to obtain donor family consent for organ donation and theatre preparation. The current recommendations regarding NHBD procurements for both uncontrolled and controlled donors is to limit the primary warm ischaemic time to 40 min, excluding periods of cardiopulmonary resuscitation [7–9]. The kidneys should be removed within 2 h of cardiac arrest with a background of *in situ* preservation. Pretransplant assessment has become necessary and been adopted into numerous NHBD programmes. This involves evaluation of the NHBD kidneys *in situ* and *ex vivo*, using a combination of perfusion features with and without machine preservation [10,11].

This is a presentation of three cases of renal transplants arising from ineffective *in situ* perfusion which resulted in adverse outcome.

Patients and methods

Previously all NHBD procurements at Newcastle-upon-Tyne since 1969 were carried out using a crash-retrieval technique without *in situ* perfusion [12]. The recent reintroduction of the NHBD programme at Newcastle-upon-Tyne in 1998 incorporated *in situ* perfusion with machine preservation [13].

From August 1998 to July 2003, 72 NHBD were referred to the transplant team. Ninety renal transplants were carried out with an overall discard rate of 38.9% (Fig. 1).

The method of pretransplant evaluation of NHBD kidneys by machine perfusion has been previously described [14,15].

In situ perfusion involves aortic perfusion using a DBTL catheter inserted through a femoral artery cut-down procedure following a '10 minuteno touch period' after cardiac arrest. The DBTL catheter is positioned with distal occlusive balloon at the aortic bifurcation using the 'pull-back' technique, the proximal occlusive balloon lies above the renal arteries. This provides selective aortic perfusion to the kidneys. When the DBTL catheter is positioned with occlusive balloons inflated, streptokinase preflush (1.5 Mega IU in 10 ml) is routinely administered and this is followed by continuous perfusion of cold heparinized Marshall's solution (1000 IU/l) [16]. The *in situ* perfusion should have a flow rate of 250 ml/min for the initial 5.0–10.0 l of perfusion fluid, from which the flow rate can then be reduced [2,5,17]. In addition our centre utilizes a roller pump to provide an adjustable peristaltic flow.

Venous decompression using a femoral vein catheter enables a 'two-way infusion system' (inflow and drainage) allowing a better perfusion and cooling technique than 'one-way infusion system' (inflow but no drainage).

The position of the DBTL catheter is approximated by measuring its anatomical location using surface anatomy (bifurcation of aorta is depicted at the umbilicus) Radiological confirmation is usually not feasible in a busy accident and emergency (A & E) department.

The time difference between *in situ* perfusion to donor nephrectomy is usually restricted to within 2 h of cardiac arrest. Therefore if *in situ* perfusion is ineffective, the viability of the NHBD kidney is compromised.

Failures of *in situ* perfusion if often seen in retrospect when eventual laparotomy reveals poor quality of *in situ* perfusion. Difficulties in *in situ* perfusion are common when there is severe atheroma in the femoral artery which limits proper placement of the DBTL.

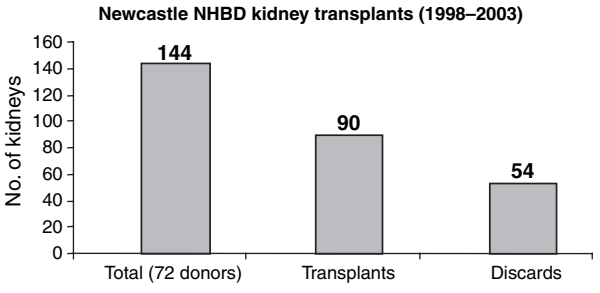

Figure 1 NHBDs, transplants and discards at the Freeman Hospital of the transplanted kidneys: Uncontrolled NHBD kidneys = 41.1%; controlled NHBD kidneys = 58.9%.

Results

The rate of an unfavourable outcome or primary nonfunction (PNF) rate from effective *in situ* perfusion was 4.8% (4/83) as opposed to 66.7% from ineffective *in situ* perfusion secondary to cannulation difficulties (Table 1). Ineffective *in situ* perfusion appears to be an indicator of PNF (chi-squared, $P < 0.0001$).

Of the ineffective *in situ* perfusion there were three donors from which a single kidney was transplanted while the contralateral kidney was discarded. The cases are

Table 1. Outcome of *in situ* perfusion (2 × 2 contingency table).

	Favourable	Unfavourable
No cannulation difficulty	83	4
Cannulation difficulty	1	2

Unfavourable outcome = primary nonfunction of allograft; favourable outcome = functional allograft.
Chi-squared test, $P < 0.0001$ for ineffective *in situ* perfusion and PNF.

Table 2. Perfusion characteristics of three NHBD renal transplants of ineffective *in situ* perfusion.

	Case 1 DG	Case 2 DI	Case 3 MZ
Donor age (years)/sex	65/M	49/M	55/M
Recipient age (years)/sex	46/M	61/M	26/M
Maastricht donor category	II	III	II
Max flow (ml/min per 100 g)	61.7	30.29	22.8
Max PFI (ml/min/100 g/mmHg)	0.696	0.59	0.53
Max tGST (IU/l/100 g)	471.8	52.53	70.00
Primary warm ischaemic time (min)	15	25	24
Secondary warm ischaemic time (min)	32	46	25
Cold ischaemic time	24 h 42 min	23 h 17 min	24 h 20 min

characterized chronologically and their ischaemic times and perfusion characteristics are illustrated in Table 2.

Case 1 (DG)

A 65-year-old man was referred to the Transplant Team as a Maastricht Category II NHBD following unsuccessful resuscitation for cardiac arrest in the A & E Department. The donor had a strong history of cardiovascular disease with previous myocardial infarctions.

An initial attempt at right femoral artery cannulation proved to be difficult, and the contralateral femoral artery was explored. As both femoral arteries were similarly affected with severe atherosclerotic disease, the DBTL catheter (14 G) was inserted partially through the right side, and the left femoral artery was tied off. *In situ* perfusion was carried out with the donor in a semi-recumbent position (45° angle head up) (M. Nicholson, personal communication).

At laparotomy the kidneys were found to be mottled and blue, which improved with machine perfusion. Both kidneys were evaluated on machine perfusion using the Newcastle hypothermic preservation system [15,18]. The left kidney was discarded, while the right kidney was transplanted into a 46-year-old male recipient with end-stage renal failure (ESRF) secondary to chronic pyelonephritis.

The perfusate glutathione S-transferase (GST) was mistakenly thought to be normal due to an error in the enzyme assay[19]. Early transplant biopsies demonstrated severe acute tubular necrosis (ATN) with 50% parenchymal infarction, the kidney eventually worked after a delayed graft function (DGF) of 26 days (Fig. 2a). He developed an adequate renal function to remain dialysis free with a stable serum creatinine of 525 μmol/l (or creatinine clearance of 16.5 ml/min).

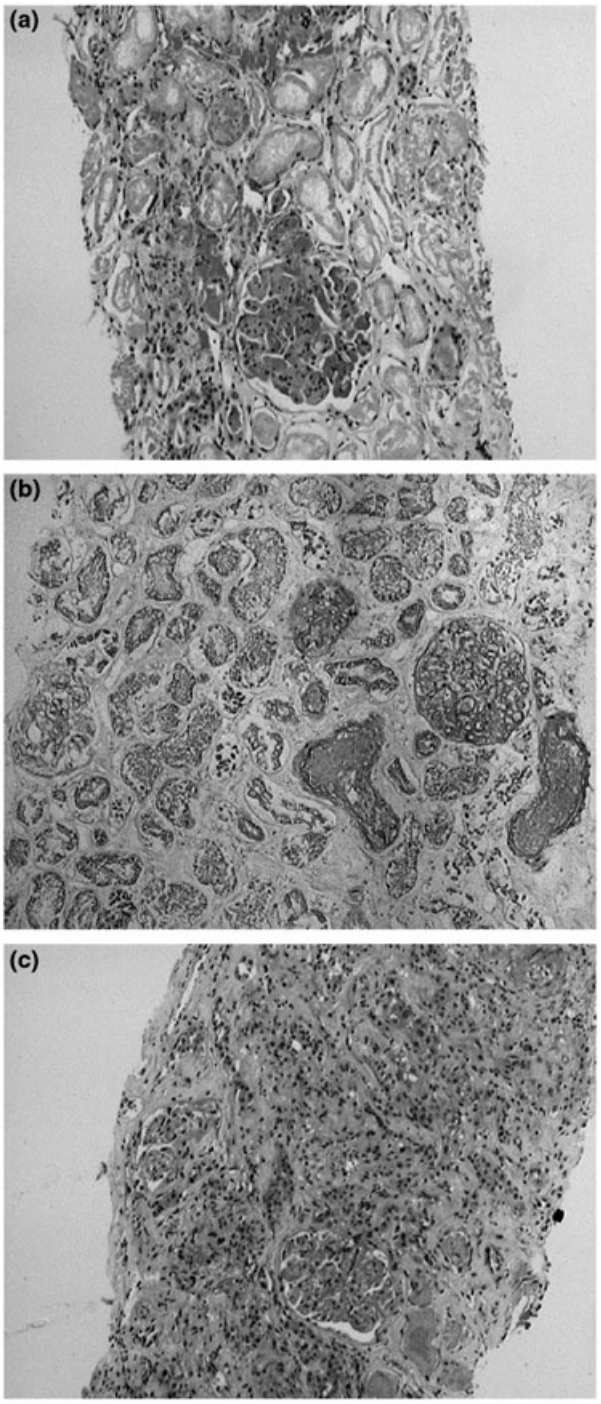

Figure 2 (a) Histology of early transplant biospy within 1 week postoperative for case 1 (×100 magnification). Histology showing approximately 50% necrosis. (b) Histology of early transplant biospy within 1 week postoperative for case 2 (×100 magnification). Histology showing approximately 50% necrosis. (c) Histology of early transplant biospy within 1 week postoperative for case 3 (×100 magnification). Histology showing approximately 50% necrosis.

Case 2 (DI)

A 49 year old male donor was referred to the Transplant Team as Maastricht category III NHBD following ''withdrawal of treatment'' in intensive therapy unit (ITU). The donor had cardiovascular disease and previous myocardial infarctions. In this case death from cardiac arrest was not anticipated immediately, therefore cannulation was carried out in ITU. Femoral cannulation was found to be extremely difficult on both sides. The DBTL catheter was inserted incompletely into the right femoral artery with the left femoral artery tied off and the patient sat up. Kidneys were exposed at laparotomy the kidneys were both blue and mottled. Both kidneys were machine perfused for 4 h. The left kidney performed poorly on machine perfusion and was therefore discarded, while the right kidney performed better on machine perfusion and was implanted into a 61-year-old man with ESRF of unknown aetiology.

The kidney never functioned with repeated early transplant biopsies demonstrating severe cortical and medullary infarction (Fig. 2b). The kidney was ultimately removed on day 30 when the recipient was showing signs of sepsis.

Case 3 (MZ)

A 55-year-old potential organ donor was identified and referred by A & E staff during unsuccessful resuscitation for cardiac arrest.

Cannulation was difficult, both sides explored and *in situ* perfused upwards with body tilt at 45° (head up).

Both kidneys appeared blue and mottled at nephrectomy, and pale after machine perfusion. The left kidney performed poorly on machine perfusion and was discarded while the right kidney was transplanted into a 26-year-old male with ESRF of unknown aetiology with a previous failed transplant.

The kidney allograft had increased his urine output but the recipient remained dialysis dependent. His dialysis sessions were reduced to two sessions per week compared with three sessions pretransplant. The early transplant biopsies demonstrated up to 50% infarction and severe ATN in adjacent areas (Fig. 2c).

Discussion

These three cases of cadaveric NHBD renal transplants highlighted the inadequacy of aortic perfusion secondary to cannulation difficulties. Attempts to correct this imperfect aortic perfusion by ligation of the contralateral femoral artery and sitting the patient up, remains difficult to assess. The kidneys procured were all blue and mottled in appearance with fair *ex vivo* perfusion. However, they were borderline passes in respect to machine perfusion characteristics and their outcome following transplantation was poor, i.e. developing PNF in two of three cases (chi-squared, $P < 0.0001$). The early postoperative biopsies showed features of severe necrosis and with infarction, demonstrating the effect of prolonged warm ischaemia of ineffective *in situ* perfusion. It therefore means that we should employ strict criteria in NHBD kidney selection, and exclude donors where insertion of the DBTL is suboptimal. The ischaemic insult in the NHBD occurs predominantly in the primary warm period, and where *in situ* perfusion is ineffective then the primary ischaemic period becomes prolonged until the kidneys become removed.

Failures in aortic *in situ* perfusion of the NHBD can be characterized as patient and procedure-dependent factors. Donor factors include: ectopic arterial anatomy or arteriosclerotic occlusive disease (i.e. iliac, aorta and renal arteries), presence of arterial stenosis (especially renal arteries), and leaks from aneurysms (aortic and iliacs).

Technical factors include: bursting of balloons, arterial inflow obstruction (kinking of cannula), venous outflow obstruction, displacement of arterial and venous catheters (insecure placement), perforation leaks and wrong-sized catheter (especially when too large diameter is used), air in tubing (tubing not primed with preservation solution) and misplacement (too high or too low).

The efficiency of *in situ* intravascular cooling with a DBTL could be cautiously evaluated before nephrectomy by cooling of the flanks (as compared with chest wall), clearing of venous outflow, and temperature fall of venous outflow. With kidneys exposed at laparotomy they are expected too be pale, soft and cool, and *ex vivo* perfusion on the back table should be easy with clear effluent.

Adequate *in situ* perfusion requires an early insertion of the DBTL catheter with restriction to lack of adequate cardiopulmonary resuscitation to <40 mins, correct uncomplicated placement of an appropriate-sized catheter, infusion tubing primed with preservation fluid, and a two-way infusion system.

Additional initiatives have been used to facilitate *in situ* perfusion including the use of streptokinase preflush, heparin, alpha-adrenergic blockers (phentolamine), the choice of *in situ* flush solution (e.g. HTK), pre-emptive insertion of the DBTL before cardiac arrest.

Viability testing with machine perfusion and enzyme analysis though useful, in these cases had failed. This may be because the perfusion was enhanced by the machine preservation system and cell disruption had not occurred producing high level of enzymes. Instead the renal cells may have been terminally injured by the period of relatively 'warm' preservation but still had intact cell membranes.

Reperfusion syndrome in these cases was presumably the last 'flow' to already fragile cells.

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