Comparison of the outcome of kidney transplantation after pulsatile or continuous ex vivo hypothermic machine perfusion of kidneys donated after cardiac death: analysis of kidney pairs

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Abstract

Background

Hypothermic machine perfusion is used to improve renal perfusion and reduce the rate of early and late graft dysfunction. It has been used in our unit since 2001. It has two modes of flow: continuous or pulsatile. The aim of this study is to compare the modes of perfusion in terms of perfusion-related parameters, graft survival and estimated glomerular filtration rate.

Methods

All donation after cardiac death kidneys between 2002 and 2014 were reviewed. Sixty-four pairs of kidneys were identified of which one kidney underwent pulsatile and the other continuous perfusion. Machine parameters including resistance and perfusion flow index levels at 0, 1, 2, 3, 4 hours were recorded and glutathione S-transferase measured in perfusate. Delayed graft function frequency, estimated glomerular filtration rate from the 1st week of transplantation until 5th year and graft survival rates were determined.

Results

Machine parameters were similar at all time points. Delayed graft function frequency, estimated glomerular filtration rates and graft survival were equivalent irrespective of perfusion mode.

Conclusion

Pulsatile perfusion may be regarded as more physiological. However, we could not identify differences in short or long term outcomes following transplantation of kidneys from the same donor that had been perfused under pulsatile or continuous conditions.

Introduction

The burden of chronic kidney disease (CKD) is increasing steadily worldwide. CKD was the 27th most common cause of death in 1990 but by 2010 it was the 18th most common [1]. More than 2 million people are now treated for kidney failure, either by dialysis or transplantation [2]. Transplantation offers superior survival and quality of life compared with dialysis [3]. Unfortunately, demand for transplantation outweighs supply of donor organs, with long waiting times for transplantation. To overcome this issue, there has been increasing use of organs donated after cardiac death (DCD) which now represents an important source of kidneys in many countries including the United Kingdom. In the UK, the number of DCD kidney transplants was 37 in 2000 whereas this increased to 619 in 2017 [4, 5].

The challenge with DCD kidneys is minimizing ischemia reperfusion injury induced by longer warm ischemia times (WIT). Ex vivo machine perfusion has been extensively investigated and used as an intervention to reduce the impact of ischemia and therefore reduce the severity of reperfusion injury, which may be particularly important for DCD kidneys. There are two main types of machine perfusion: normothermic and hypothermic perfusion. Normothermic machine perfusion is a more recent development and its benefit is still being evaluated [6]. In contrast, hypothermic perfusion has been evaluated in several clinical trials. Moers et al showed that machine perfusion is superior to static cold storage improving 1 year graft survival and reducing the duration of delayed graft function [7, 8]. There are other studies reporting an advantage of machine perfusion over static cold storage [9-13]. Hypothermic perfusion has even been used for living donor kidneys with estimated glomerular filtration rate (eGFR) higher at 1 year in recipients of kidneys that had undergone hypothermic machine perfusion compared to static cold storage [14]. Two types of hypothermic perfusion machines are available: the RM3 Renal Preservation System (Waters Medical Systems) and the Lifeport Kidney Transporter (Organ Recovery Systems). Lifeport machine perfusion has been used in Newcastle upon Tyne since 2001. The latter has two modes of perfusion, continuous or pulsatile. The pulsatile mode perfuses kidney at a set

systolic pressure 30 times in a minute. The continuous mode perfuses kidneys at a constant pressure [15].

There is limited data comparing the modes of perfusion. To the best of our knowledge, the effects of the different modes of perfusion available on the Lifeport machine on transplant outcome have not been compared. The aim of this study is to assess the effect of pulsatile and continuous machine perfusion modes on eGFR and graft survival, uniquely using pairs of kidneys from the same donor.

Material and Methods

When two Lifeport machines were purchased from Organ Recovery Systems, the perfusion mode was set as pulsatile for one machine and continuous for the second. Therefore, for DCD donors providing two kidneys, each kidney had a different mode of perfusion. All DCD kidney transplants between 2002 and 2014 at the Freeman Hospital, Newcastle upon Tyne NHS Foundation Trust were retrospectively reviewed. Exclusion criteria were patients receiving dual transplants, kidneys without machine perfusion, kidneys in which one of the pair was transplanted at another center, kidney pairs where both perfused on same mode. Figure 1 summarizes details of the two groups. Details about donor's age, sex, Maastricht category, presence of donor hypertension, 1st and 2nd warm ischemia time, cold ischemia time (CIT) were collected from donor request forms and clinical notes. Details of recipient's sex, age, transplantation date, graft survival details, creatinine values, presence of delayed graft function (DGF) and eGFRs at 1st week, 1st, 3rd, 6th, 9th, 12th months, 2nd, 3rd, 4th, 5th years were collected from hospital record systems. DGF was defined as the requirement for dialysis in the first 7 days after transplantation.

Machine perfusion data including machine type, perfusion flow index (PFI), resistance (RT) [16], and GST glutathione S-transferase (GST) concentrations were collected. During perfusion, systolic pressure was increased to 30 mm Hg and fixed at that pressure. PFI is calculated by dividing flow by systolic pressure and expressed as mL/min/mm Hg per 100 g of kidney. RT is calculated as mean pressure divided by flow at specified time and presented as mm Hg/mL/min. Total GST level were measured

from the perfusate fluid. Perfusion measurements and 10 mL perfusate for GST analysis were taken at 0, 1st, 2nd, 3rd, 4th hour of perfusion. Belzer UW machine solution was used for both modes of perfusion. All patients had received induction with basiliximab and standard triple maintenance immunosupression consisting of tacrolimus, mycophenalate mofetil and prednisolone as regulated by Freeman Hospital guideline. The target trough tacrolimus levels were 6-10 µg/l for the first sixth months, 5-8 µg/l for the second sixth months and 3-7 µg/l thereafter.

Statistical Analysis

Statistical analysis was performed by Scientific Package for Social Science (version 15.0; SPSS, Chicago, Illinois). Continuous variables were compared by independent samples t test and Kruskall-Wallis test. Categorical variables were compared by Chi-square test. Graft survival was analysed by Kaplan-Meier curves and log-rank method.

Results

Demographic data

In total, data from 241 kidneys transplanted between 2002 and 2014 were analysed. After exclusion of 113 kidneys for reasons detailed Figure 1, 64 pairs of kidneys were included of which one of the pair was perfused in pulsatile mode and the other was perfused in continuous mode.

Demographic characteristics of donors and recipients, details about ischemic times are given in Table 1. Pulsatile and continuous groups were similar in comparison of recipient age, 1st and 2nd WIT, total WIT and CIT ($p>0.05$ for all). Female recipients were 21.9% of the pulsatile group and 39.1% of the continuous group ($p=0.035$). Donor age, hypertension, Maastricht category were identical as pairs of kidneys were analysed.

The effect of machine mode on perfusion parameters and outcome

A summary of all perfusion parameters is given in Table 2. RT values decreased gradually in both groups and comparison showed they were similar ($p>0.05$). GST values from 0 hour until 4 hour were also similar between pulsatile and continuous mode ($p>0.05$ for all analyses). Although PFI values were all higher in continuous mode for all times, these did not differ significantly from pulsatile mode ($p>0.05$ for all). DGF occurred in 22.8% of transplants following pulsatile perfusion and 25.4 % following

continuous perfusion (p=0.742). Estimated glomerular filtration rates between pulsatile and continuous groups were similar from just after transplantation (1 week) until the 5th year (p>0.05 for all time points) (Figure 2). Graft survival was also similar between two modes of perfusion (log rank p =0.803) (Figure 3).

Discussion

This study evaluated the possible relationship between machine mode during hypothermic perfusion and long term outcomes of DCD kidney transplants. Selection of the study population to contain only pairs of kidneys excluded many possible biases in this study. Perfusing one kidney on pulsatile and the other kidney of the pair in continuous mode made donor characteristics including donor age, donor sex, donor hypertension, Maastricht Category identical in the two groups. Ischemic times were also similar between groups. Recipients received the same induction and standard triple maintenance immunosupression according to local guidelines. Therefore any difference in outcome is likely to be due to the mode used for hypothermic perfusion.

There are some studies comparing static cold storage with hypothermic perfusion using the Lifeport machine without citing mode of perfusion [9, 10, 13], with continuous mode [11] or with pulsatile mode [12]. These studies showed benefit of machine perfusion over static cold storage in terms of recovery of renal function and length of hospital stay [9] , graft survival [10, 12], delayed graft function [11, 13], although a benefit has not been seen in all studies [17]. In addition, Kozaki et al reported their experience of kidneys perfused on continuous mode to define viability of donated kidneys. [18]. Two meta analyses have been performed comparing hypothermic machine perfusion to static cold storage [19, 20]. Machine perfusion had short term benefits with decreased rate of DGF. Long term results were similar. However, these studies did not address the question of whether continuous or pulsatile perfusion is superior.

Perfusion modes have been compared in two animal studies. Implantation of left ventricular assistance devices was used to measure the effects of pulsatile and continuous systemic perfusion on renal sympathetic nerve activity in Mongrel dogs [21]. Sympathetic nerve activity and peripheral vascular resistance were decreased by pulsatile perfusion. The authors concluded that pulsatile perfusion may

therefore improve both microcirculation and organ function. Lindell et al compared modes of Lifeport machine perfusion Beagle dogs [22]. After a warm ischemia period for 45 minutes, the left kidneys of 12 dogs were removed. Eight kidneys were maintained by pulsatile machine perfusion and 4 by continuous perfusion for 24 hours. Kidneys were autotransplanted after machine perfusion at which stage the right kidneys were removed. Serum creatinine levels were significantly lower in the pulsatile group. The survival was 50% in the continuous group and 100% in the pulsatile perfusion group.

This current study is the first to compare the two perfusion modes in clinical transplantation. No difference in either short (DGF) or long-term transplant outcomes (EGFR and transplant survival) were identified. Although a retrospective analysis of data, the study is strengthened by the ability to compare outcomes in pairs of kidneys from the same donor, one of which underwent continuous and the other pulsatile perfusion. Therefore, any theoretical advantage of more physiological pulsatile perfusion, supported by pre-clinical data, does not translate into clinical benefit.

In summary, whatever the mode of perfusion, machine perfusion has been shown to be superior to static cold storage in many studies. Animal studies comparing perfusion modes favor pulsatile perfusion. Pulsatile perfusion is perhaps more physiological, but despite this, we did not identify any difference in resistance or PFI between pulsatile of continuous flow. GST concentrations in the perfusate, as a biomarker of renal cell injury, were similar in kidneys undergoing pulsatile or continuous perfusion. This suggests that non-physiological continuous perfusion is not causing pressure-related injury to the perfused kidney. Most importantly, and in contrast to animal studies, we have found similar graft survival rates and eGFR values between different modes of perfusion.

Conclusion

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This is the first study comparing the mode of hypothermic machine perfusion in human kidneys. Results of this study reveal that machine perfusion modes are not superior to each other, with similar DGF rates, graft function and survival when the two modes are compared.

Keywords: Continuous machine perfusion, donation after cardiac death, hypothermic machine perfusion, pulsatile machine perfusion

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Figure Legends

Figure 1. Details of patient cohort.

All donation after cardiac death (DCD) kidney transplantations from 2002-2014 were included.

Figure 2. eGFR values according to machine mode.

The solid line represents pulsatile mode, dotted line represents continuous mode. eGFR was similar at all time points ($p>0.05$ for all).

Figure 3. Comparison of graft survival according to perfusion mode.

Kaplan-Meier analysis of graft survival following pulsatile (solid line) or continuous (dotted line) hypothermic machine perfusion ($p=0.803$). Data is censored for death with a functioning graft.

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