Transpl Int (1996) 9 [Suppl 1]: S452–S454

J. A. B. van der Hoeven
I. J. de Jong
R. F. E. Wolf
R. L. Kamman
R. J. Ploeg

Tissue hydration in kidneys during preservation: a relaxometric analysis of time-dependent differences between cortex and medulla

J. A. B. van der Hoeven (✉) · I. J. de Jong · R. J. Ploeg
Department of Surgery,
University Hospital Groningen,
Hanzeplein I, PO Box 30.001,
NL-9700 RB Groningen,
The Netherlands

R. F. E. Wolf · R. L. Kamman
Department of Radiology,
University Hospital Groningen,
Hanzeplein 1, PO Box 30.001,
NL-9700 RB Groningen,
The Netherlands

Abstract Cold preservation of donor organs induces hypothermia-related tissue edema as a result of a reduced activity of the ATP-dependent sodium pump at low temperatures. Hypothermia-induced tissue edema occurs in kidney preservation and is a significant risk factor for delayed graft function (DGF) after transplantation. DGF remains a major problem in kidney transplantation and is significantly associated with preservation injury. The state of hydration of cold-stored organs can be assessed from a biopsy for determination of the wet/dry weight ratio. As a non-invasive method to determine tissue hydration MRI T_1 and T_2 relaxometry can be used. In this study we have compared changes in tissue hydration in UW-preserved porcine kidneys with increasing cold ischemia times (CIT) using wet/dry weight ratio and MR ralaxometry. The results of the two techniques were correlated to evaluate the use of MR relaxometry. Wet/dry weight ratios of the renal cortex decreased with prolonged CIT ($P < 0.01$) whereas those of the medulla did not change significantly. T_1 values of the cortex decreased with prolonged CIT ($P < 0.01$). T_2 values of the cortex showed a non-significant decline with increased CIT. No significant changes in T_1 and T_2 were found in the medulla. The correlation between T_1 and the wet/dry weight ratio of the cortex was significant ($P = 0.05$, linear correlation coefficient 0.8698). We conclude that MR relaxometry can be a valuable non-invasive technique to assess tissue hydration in cadaveric donor kidneys before transplantation.

Key words Preservation · Kidney · Tissue hydration, MRI

Introduction

During cold-storage preservation of donor organs reduced activity of the ATPase-dependent sodium pump causes sodium and water influx into the cell [1, 2]. With longer cold ischemia times (CIT) more edema in the tissue is expected. The hypothermia-induced tissue edema is a potential factor contributing to the incidence of delayed graft function (DGF) after kidney transplantation. DGF remains a major problem in kidney transplantation and is associated with preservation injury [3, 4, 6]. The state of hydration of the cold-stored organ can be determined with a surgical biopsy. The difference between the wet weight and the dry weight of the biopsy represents the hydration state of the tissue, the wet/dry weight ratio. MRI offers a non-invasive alternative method to assess tissue hydration by measuring the T_1 and T_2 relaxometric characteristics. In previous reports, MR relaxometry has been used to evaluate changes in tissue hydration of ischemic reperfused kidneys in an experimental model and in human cold-stored donor livers [5, 6]. In this study we have looked at the changes in tissue edema in UW-preserved porcine kidneys with increasing CIT. MR relaxometry was compared to the standard wet/dry weight ratios.

Table 1 Wet/dry weight ratio and T_1 and T_2 relaxometric data. Data are shown as means ± SD ($n = 20$) per cold ischemia time *(CIT)* group

	6 h CIT	12 h CIT	24 h CIT	36 h CIT	72 h CIT	*P* values
Mean T_1 cortex	491.56 ± 28.00	474.20 ± 27.37	470.85 ± 31.50	460.93 ± 27.63	463.59 ± 28.64	< 0.01
Mean T_2 cortex	64.22 ± 5.55	63.79 ± 6.18	62.62 ± 6.67	61.97 ± 5.46	60.63 ± 4.74	NS
Mean T_1 medulla	745.32 ± 54.31	738.62 ± 52.72	734.20 ± 57.53	712.03 ± 48.06	739.04 ± 38.64	NS
Mean T_2 medulla	105.89 ± 13.43	108.93 ± 16.58	104.61 ± 17.80	104.97 ± 14.76	105.69 ± 10.39	NS
Wet/dry weight ratio cortex	0.8058 ± 0.009	0.8018 ± 0.010	0.7878 ± 0.013	0.7829 ± 0.011	0.7752 ± 0.010	< 0.01
Wet/dry weight ratio medulla	0.8553 ± 0.022	0.8621 ± 0.009	0.8595 ± 0.007	0.8479 ± 0.008	0.8579 ± 0.013	NS

Materials and methods

Twenty porcine kidneys were harvested from 6-month-old female Dutch pigs. After a warm ischemic period of 20 min, the kidneys were flushed with 250 ml of UW solution (0–4 °C) on a backtable. After the UW flush, the kidneys were divided into a caudal and a cranial part. Both parts were stored in UW-filled dishes and placed in seperate ice-filled styrofoam containers. At 6, 12, 24, 36, and 72 h CIT, MR relaxometry was performed on the cranial segments of the kidneys. At the same time, biopsies were taken from the cortex and the medulla of the caudal segments for wet/dry weight radio determination.

Wet/dry weight ratio

The surgical biopsies were taken from the caudal segment of the kidneys. Both medullary and cortical biopsies were obtained in a room with constant relative humidity. The biopsies were blotted twice on blotting paper before they were weighted. After the wet weight was established, the biopsies were stored in biofreeze vials (Co-Star 2 ml) in liquid nitrogen. The dry weight was determined after dry freezing for 120 h. To ascertain that the tissue was completely dry, five samples were freeze-dried for another 120 h in which they did not loose additional weight. Then the wet/dry weight ratio was calculated.

MRI and relaxometry

Images were obtained with a 1.5 Tesla whole-body system (Gyroscan S15/HP, Philips Medical Systems, Best, The Netherlands). Scoutview image allowed exact determination of the kidneys in the container and also allowed planning of the relaxometric imaging series. Images were obtained using a 'mixed pulse sequence' which is a combination of a spin echo (SE) and an inversion recovery (IR) pulse sequence. Both sequences were applied with eight echos and echo times of 30 msec. One average was made of a single 5-mm-thick slice with a scan matrix resolution of 256 × 256 in a 500 mm field of view. The repetition time for the SE sequence was 100 msec and for the IR sequence 1400 msec, with an inversion time of 400 msec. With standard system software features, images with signal intensities directly proportional to T_1 and T_2 (relaxometric images) were calculated. The two-dimensional fast Fourier technique (2D-FFT) was used for image reconstruction.

Image analysis

Reconstructed images were presented in a 256 × 256 pixel matrix and studied on a viewing console (Philips Medical Systems, Best, The Netherlands). Relaxation parameters, T_1 and T_2, were calculated as follows: in every calculated T_1 and T_2 image six regions of interest (ROI) of 0.1 cm^2 were positioned, three ROI in the cortex and three ROI in the medulla. The ROI were positioned in such a way that the contribution of UW in the vessels was minimal and there was no partial volume effect. In this way, information about T_1 and T_2 characteristics was obtained from 0.05 cm^3 renal parenchyma with each ROI. A mean T_1 and T_2 of cortex and medulla was calculated for each kidney.

Data analysis

Statistical analysis was performed using the paired Student's *t*-test and a linear correlation coefficient with $P < 0.05$ considered significant.

Results

The results of the determination of the tissue hydration state with wet/dry weight ratios and T_1 and T_2 relaxometry are shown in Table 1. The wet/dry weight ratio of the renal cortex decreased significantly from a mean of 0.8058 at 6 h CIT to a mean of 0.7752 at 72 h CIT ($P < 0.01$, paired Student's *t*-test). The wet/dry weight ratios of the renal medulla did not change during the same period. The MR relaxometric data showed a significant decline of T_1 and T_2 values of the cortex with longer CIT from 6 to 72 h ($P < 0.01$, paired Student's *t*-test). No significant changes in T_1 and T_2 were measured in the medulla although T_1 declined. The correlation between T_1 and the wet/dry weight ratio of the cortex was significant (linear correlation coefficient 0.8698, $P < 0.05$) (Table 1).

Discussion

The decrease in the wet/dry weight ratio of the cortex demonstrates the dehydrating properties of UW solution due to the impermeants lactobionate and raffinose. In our model, UW solution protected against the development of tissue edema in the medulla since the wet/dry weight ratio did not change. The dehydrating properties of the UW solution have been reported to exceed those of Eurocollins or phosphate-buffered sucrose in experimental models [5]. A significant correlation was

seen between MR relaxometric data and wet/dry weight ratios. This indicates that MR relaxometry could be a valuable non-invasive alternative technique to assess tissue hydration in cadaveric donor kidneys during cold storage. Further studies are in progress to investigate the clinical value of MR relaxometry in human cadaveric donor kidneys and its relation to the incidence of posttransplant delayed graft function.

References

1. Belzer FO, Southard JH (1988) Principles of solid organ preservation by cold storage. Transplantation 45: 673–676
2. Martin DR, Scott MDF, Downes GL, Belzer FO (1972) Primary cause of unsuccesful liver and heart preservation: cold sensitivity of the ATPase system. Ann Surg 175: 111
3. Ploeg RJ, Bockel H van, Langendijk PTH, et al (1992) A randomized controlled clinical trial comparing preservation with UW solution and Eurocollins in cadaveric renal transplantation. Lancet 340: 129–137
4. Thiel G et al (1982) The role of reduced medullary perfusion in the genesis of acute ischaemic renal falure. Nephron 31: 321
5. Walhberg JA, Southard JH, Belzer FO (1986) Development of a cold storage solution for pancreas preservation. Cryobiology 23: 477
6. Yamamoto K, Wilson DR, Baumal R (1984) Outer medullary circulatory defect in ischaemic acute renal failure. Am J Pathol 116: 253