MicroMR Imaging of Renal Interstitial Fibrosis in a Unilateral Ureteric Obstruction Model

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TARGET AUDIENCE: Nephrologists, DCE-MRI practitioners

PURPOSE: Renal fibrosis is the final common pathway of renal injury and results in chronic renal failure. There is a need for a non-invasive method to quantify renal fibrosis so as to study the severity of disease progress and the efficacy of anti-fibrotic therapy. DCE-MRI has been shown to be able to measure GFR non-invasively but has not been investigated as a biomarker for renal fibrosis. DWI has been shown to correlate with renal fibrosis.[1] Renal fibrosis is induced by unilateral ureteric obstruction (UUO) with survival surgery. We aim to determine if a tracer kinetic model[2] applied to DCE-MRI and combined with DWI can serve as a biomarker for renal fibrosis.

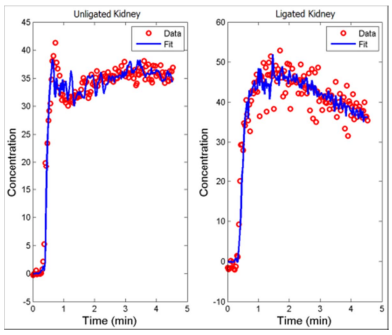

Fig 1. Typical DCE signal concentration time curves at D7 after UUO in the unligated and ligated kidney

METHODS

Mice: The study was approved by the local institutional animal care and use committee. Six male C57/BL6 mice (8 weeks old, 20-30g) were maintained according to the Guide for the Care & Use of Laboratory Animals (NIH). The mice were scanned before the UUO procedure (day -1) and on day 7 after the UUO.

UUO: A midline abdominal incision was made for laparotomy. 8/0 monofilament suture (J and J, non absorbably nylon) was placed around the ureter and a surgical knot was tied to obstruct the ureter.

MRI: MRI was performed on a 7T scanner (Bruker ClinScan, Bruker BioSpin MRI GmbH, Germany). For DWI, a multishot spin-echo EPI sequence was used with following parameters: TR = 3000 ms, TE = 41 ms, FOV = 36 x 36 mm, 64 x 64 matrix, 8 slices with thickness of 1 mm, and *b* values of 0, 50, 100, 200, 400, 800, 1200 s/mm^2. For DCE imaging, a 3D VIBE sequence was used with following parameters: TR = 3.04 ms, TE = 1.23 ms, FOV = 36 × 36 mm, 128 × 128 matrix, 8 slices with thickness of 1 mm, & temporal resolution 2 s. Five sets of baseline images were acquired with α = 6° & 14°. It was followed by a dynamic sequence of 130 sets of images (α = 14°). 100 µL of Gd-DOTA (Dotarem, Guerbet SA, France) was manually injected through the tail vein after the first set of dynamic images and followed by flushing with 200 µL of saline.

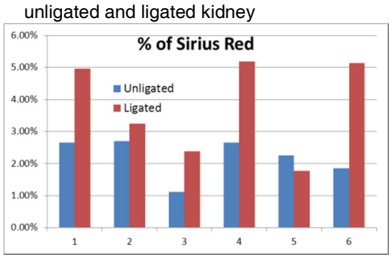

Fig 2. Percentage of Sirius Red stain area in both kidneys of each mouse, showing an elevated level of fibrosis in the ligated kidneys

Data Processing: Region of interests corresponding to the cortices of each kidney and renal artery were manually outlined. Microcirculatory parameters such as renal plasma flow (*F*), blood volume (v_b), and parenchymal mean transit time (PMTT) were derived from Fine's model.[2]

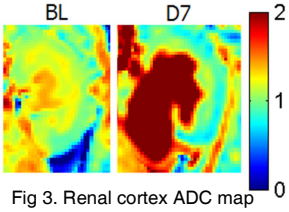

Fig 3. Renal cortex ADC map of a ligated kidney at baseline and D7 after UUO, showing a drop of ADC in the renal cortex after UUO

Histopathology: The kidneys were sectioned and stained with Sirius Red for quantification of collagen which was used as a marker of fibrosis. The images were analysed using Measure Stain Area algorithms of Slidepath TissueIA software (Leica Microsystems, Germany). Scanning and image analysis was performed at Advanced Molecular Pathology Laboratory, IMCB, Singapore.

RESULTS: Signal concentration time curves of an unligated and ligated kidney at day 7 after UUO are shown in Figure 1. There is an increase of collagen deposit in the ligated kidneys (paired t-test, p = 0.040; Fig. 2). There is a significant drop of PMTT (p = 0.011) and ADC (p = 0.013; Fig 3) at day 7 in the ligated kidneys, as shown in Table 1. There is a significant correlation between % area positively stained by Sirius Red and PMTT (p = 0.012; Fig 4).

Parameter	Unligated Kidney		Ligated Kidney	
	Baseline	D7	Baseline	D7
F (ml/min)	2.31 ± 1.40	2.86 ± 1.12	3.69 ± 1.36	3.60 ± 2.13
v_b (%)	3.91 ± 1.52	3.36 ± 1.69	3.30 ± 1.83	3.95 ± 1.82
PMTT (s)	213.36 ± 63.60	228.91 ± 28.41	205.62 ± 54.75	107.20 ± 22.60
ADC (10^{-3} mm^2/sec)	1.07 ± 0.12	1.20 ± 0.08	1.13 ± 0.08	0.97 ± 0.09

DISCUSSION: The drop of ADC in the ligated kidneys is concordant with the findings of Togao.[1] PMTT is strongly linked with renal function[3] and the washout pattern seen in the ligated kidneys (Fig 2), which yielded low values of PMTT, indicated the deterioration of renal function in the ligated kidneys. The significant correlation observed between PMTT and degree of fibrosis as measured by % of Sirius Red showed potential of using DCE-MRI as biomarker of renal fibrosis.

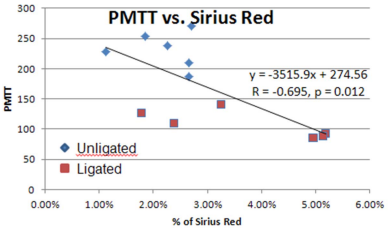

Fig 4. Correlation between PMTT and percentage of Sirius Red stain area in each kidney

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