

# T1rho MR imaging for rat liver with carbon tetrachloride intoxication: a time course study

Feng Zhao<sup>1</sup>, Min Deng<sup>1</sup>, Jing Yuan<sup>1</sup>, Queenie Chan<sup>2</sup>, Jun Yu<sup>3</sup>, and Yi-Xiang Wang<sup>1</sup>

<sup>1</sup>Department of Imaging and Interventional Radiology, The Chinese University of Hong Kong, Shatin, N.T., Hong Kong, <sup>2</sup>Philips Healthcare, Hong Kong, <sup>3</sup>Department of Medicine and therapeutics, The Chinese University of Hong Kong, Shatin, N.T., Hong Kong

**Introduction:** By performing T1 rho MR imaging, it was shown that the relaxation parameter, T1 rho, became prolonged after bile duct ligation (BDL)-induced liver injury in an animal model and that the T1 rho value increased with the severity of liver fibrosis [1], therefore liver MRI T1rho value might be an important biomarker for liver fibrosis [1, 2]. A few important questions remain to be further answered: 1) Is MR T1rho sensitive to liver fibrosis with an etiology other than biliary fibrosis? 2) How liver acute inflammation and edema will affect liver T1rho value? To answer these questions, the current experimental study was carried out using rat liver fibrosis model induced with chronic carbon tetrachloride (CCl4) insult. This animal model has been clearly characterized and in many respects mirrors the pattern of disease seen in human fibrosis and cirrhosis associated with toxic damage [3].

**Material and Methods:** The protocols and procedures were approved by the local Animal Experimentation Ethics Committee. Male Sprague-Dawley rats with weight of 200-250g were used. For liver fibrosis induction, the rats were administered 1:1 volume mixture of carbon tetrachloride (CCl4,99.8%; BDH Laboratory) in olive oil. Liver fibrosis in rats was induced by intraperitoneal injection of 2 mL kg-1 body weight CCl4 twice weekly up to 6 weeks. All rats were allowed free access to water and diet, and weighed twice weekly. Data acquisition was performed on a 3T clinical scanner (Achieva, Philips Healthcare, Best, The Netherlands). After anesthesia, animals were positioned supine and an 8-channel human wrist radiofrequency (RF) coil was used as the signal receiver, and the in-built body coil was used as the signal transmitter. Five axial slices were selected to cut through liver. For T1rho measurement, a rotary echo spin-lock pulse was implemented in a 3D balanced fast field echo (bFFE) sequence. Spin-lock frequency was set as 500 Hz and the spin-lock times of 1 ms, 10 ms, 20 ms, 30 ms, 40 ms, and 50 ms were used for T1p mapping. TE and TR were 2.6 ms and 5.3 ms respectively. FOV: 80×66×10mm<sup>3</sup>, voxel size: 0.50×0.50×2.00 mm<sup>3</sup>. The flip angle was 40 degree and the number of signal averages (NSA) was 4. Delay time after acquisition was set as 6000 ms to restore equilibrium magnetization prior to the next T1p preparation. Conventional T2 weighted image of rat liver were acquired by turbo spin echo (TSE) sequences with TSE factor=16, TE/TR=80ms/2000ms, voxel size=0.50×0.5×2mm<sup>3</sup>, and NSA=4. T1rho maps were computed on a pixel-by-pixel basis using a mono-exponential decay model of  $M(TSL)=M_0 \cdot \exp(-TSL/T1rho)$  with a home-made Matlab program (Mathworks, Natick, MA, USA). To quantify T1p value on T1p maps and liver signal intensity on T2 weighted images, five regions-of-interest (ROI) of approximately 3-4mm<sup>2</sup> were placed on each slice of the liver parenchyma region, leading to a total of 25 ROIs from each liver for each technique. For T2 weighted images, the mean signal intensity of liver parenchyma was normalized by the signal intensity of the back muscle on the same image, i.e. the ratio of liver signal intensity and back muscle intensity was obtained. Animals were MR imaged at baseline prior to the CCl4 injection (n=12), 48 hours post initial CCl4 injection (n=12), and 2 weeks (n=12), 4 weeks (n=10), 6 weeks (n=7) post the initiation of CCl4 injection. At each time point, three rats were killed for liver histology including standard HE staining and Picrosirius red staining (Fig1).

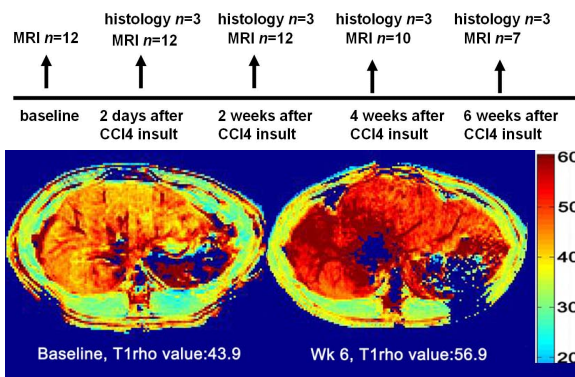


Fig 1. Schedule of CCl4 administration for the induction of liver fibrosis in adult Sprague-Dawley rats, MRI data acquisition, and liver histology.

**Results:** The rat liver baseline T1rho value was  $44.13 \pm 1.0$ ms. 48 hours post the initiation of CCl4 insult, the liver T1rho value was  $46.06 \pm 1.27$ ms, increased by 4% ( $P<0.001$ ). At week 2, week 4, and week 6, the liver T1rho value was  $49.43 \pm 3.69$ ms,  $51.43 \pm 3.79$ ms, and  $54.29 \pm 2.94$ ms respectively, with an increase over baseline value of 12% ( $P<0.001$ ), 17% ( $P<0.001$ ), and 23% ( $P<0.001$ ) respectively (Fig 2). Normalized liver signal intensity on T2 weighted image was  $0.96 \pm 0.19$  at baseline. 48 hours post the initiation of CCl4 insult, this value was  $1.32 \pm 0.23$ , increased by 37% ( $P=0.001$ ). At week 2, week 4, and week 6, liver normalized liver signal intensity on T2 weighted image was  $1.23 \pm 0.19$ ,  $1.39 \pm 0.18$ , and  $1.25 \pm 0.2$  respectively; with an increase over baseline value of 28% ( $P=0.005$ ), 44% ( $P<0.001$ ), and 31% ( $P=0.011$ ) respectively (Table 2). Histology results showed 2 days after CCl4 insult, extensive infiltration of inflammatory cells, intracellular fat vacuoles deposition and hepatocellular swelling were observed in the liver without apparent collagen deposition. At week 2, 4, and 6, in addition to infiltration of inflammation cells, intracellular fat vacuoles deposition, necrosis of hepatocytes and progressive liver fibrosis were seen.

Fig 2. Typical T1p maps of one rat at baseline before CCl4 insult and week 6 after CCl4 insult. Table 3. T1rho values and normalized liver signal intensity on T2 weighted image at different time points (Baseline: before CCl4 insult; CCl4 48 hours: after 48 hours of CCl4 insult; CCl4 Week 2,4,6: after 2,4,6 weeks of CCl4 insult).

	Baseline	CCl4 48 hours	CCl4 Week 2	CCl4 Week 4	CCl4 Week 6
T1rho	$44.13 \pm 1.0$ ms	+4% ( $P<0.001$ )	+12% ( $P<0.001$ )	+17% ( $P<0.001$ )	+23% ( $P<0.001$ )
SI*	$0.96 \pm 0.19$	+37% ( $P=0.001$ )	+28% ( $P=0.005$ )	+44% ( $P<0.001$ )	+31% ( $P=0.011$ )

SI\* Normalized liver signal intensity on T2 weighted image

**Discussion:** Compared with BDL-induced liver injury model, the CCl4 model is associated with more extent of inflammation, edema, and tissue necrosis. Liver T1rho value increased slightly 48 hours post CCl4 insult, and then increased further and was the highest at 6 weeks post insults. The contribution from acute inflammation and edema to the liver T1rho increase, as seen with 48 hours' results, seems to be mild. While edema, reflected as signal on T2 weighted images, reached a high value 48 hours post CCl4 insult, MRI T1rho value increased progressively during the course of CCl4 insults, therefore edema and T1rho changes did not follow a similar course. In conclusion, MR T1rho is sensitive to liver fibrosis due to CCl4 intoxication, and liver acute inflammation and edema do not substantially affect liver T1rho value.

**References:** [1] Wang YX, et al. Radiology. 2011;259(3):712-9. [2] Sirlin CB. Radiology. 2011;259(3):619-20. [3] Constandinou C, et al. Methods Mol Med 2005;117:237-50.