Non-invasive quantification of hepatic fibrosis in mouse models by magnetic resonance relaxometry (MRR)

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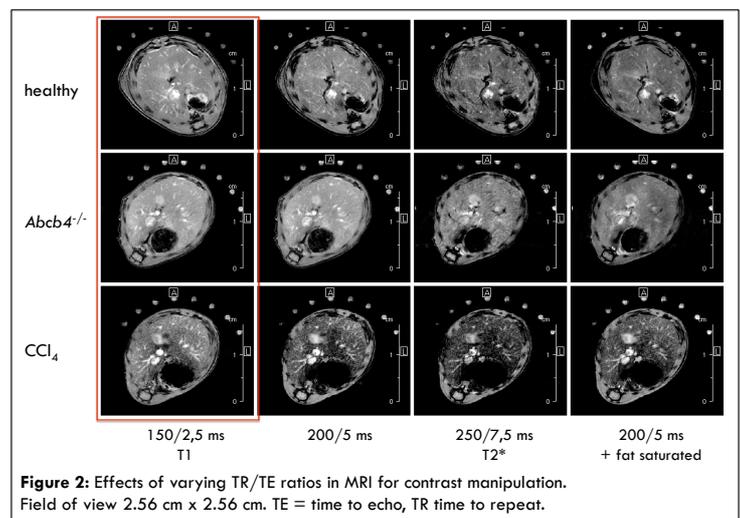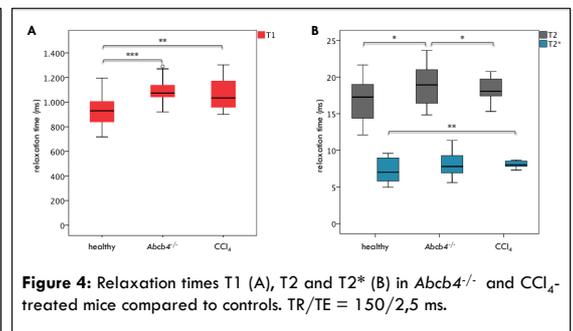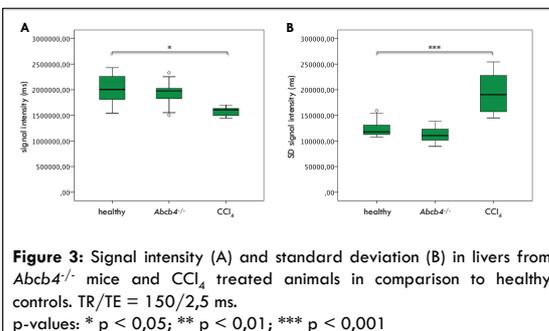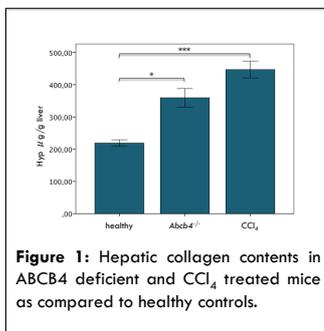[2]Department of Medicine II, Saarland University Medical Center, Homburg, Germany

Background: To date studies in mouse models of hepatic fibrogenesis have been hampered by the need for tissue specimens to quantify liver fibrosis. Hence, the development of non-invasive methods of quantifying hepatic scarring is crucial to monitor potential antifibrotic therapies.

Our aim was to develop non-invasive magnetic resonance-based quantification of liver fibrosis in mice with different aetiologies of fibrosis.

Methods: We analysed 13 BALB/cJ inbreed mice after toxic induced liver fibrosis (1.4 mg CCl_4/kg/wk i.p. for 6 wks), 27 *Abcb4* knockout (*Abcb4$^{-/-}$*) mice with biliary fibrosis, and 26 controls. To assess hepatic fibrosis, we ascertained fibrosis stages (F-scores) after Sirius red staining and hepatic collagen (hydroxyproline) contents. Magnetic resonance imaging (MRI) and relaxometry (MRR) were performed on a horizontal bore 9.4 T animal scanner (Bruker Biospec) with a circular polarized receive/transmit coil. Relaxation times T1, T2 and T2* were acquired with turbo spin and multiple gradient echo sequences. Four MRI experiments were performed, varying the spoiled gradient echo sequence from T1 to T2* weighting with signal intensity (Si) measurements.

Figure 2: Effects of varying TR/TE ratios in MRI for contrast manipulation. Field of view 2.56 cm x 2.56 cm. TE = time to echo, TR time to repeat.

Results: Compared to wild-type controls, animals treated with CCl_4 and *Abcb4$^{-/-}$* mice display significantly (p<0.01) enhanced hepatic collagen contents (Fig. 1) and F-scores (not shown). As illustrated in Fig. 2, MRI experiments demonstrate differences in tissue morphology between *Abcb4$^{-/-}$*, CCl_4 treated animals, and controls. While Si (Fig. 3A) is significantly reduced, Si deviation (Fig. 3B) is enhanced much larger in animals treated with CCl_4 as compared to healthy controls. T1 relaxation times (Fig. 4A) differ between healthy controls and *Abcb4$^{-/-}$* mice as well as CCl_4-treated animals. However, *Abcb4$^{-/-}$* mice display significantly increased T2 relaxation times (Fig. 4B) in comparison to healthy animals.

Figure 1: Hepatic collagen contents in ABCB4 deficient and CCl_4 treated mice as compared to healthy controls.

Figure 3: Signal intensity (A) and standard deviation (B) in livers from *Abcb4$^{-/-}$* mice and CCl_4 treated animals in comparison to healthy controls. TR/TE = 150/2,5 ms.
p-values: * p < 0,05; ** p < 0,01; *** p < 0,001

Figure 4: Relaxation times T1 (A), T2 and T2* (B) in *Abcb4$^{-/-}$* and CCl_4-treated mice compared to controls. TR/TE = 150/2,5 ms.

Conclusion: Our study demonstrates the feasibility of MRR as a non-invasive method to discriminate between fibrotic and non-fibrotic liver tissue in mice independent of the aetiology of hepatic injury. Additional studies are currently being performed to develop algorithms allowing the refined differentiation of fibrosis stages and the assessment of fibrosis resolution to provide a tool for the monitoring of antifibrotic therapies *in vivo*.

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