MR elastography for the early detection of steatohepatitis in the rat with fatty liver

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Introduction

The early and non-invasive detection of non-alcoholic steatohepatitis in the fatty liver is difficult. The aim of our study in the rat was to assess the potential value of magnetic resonance (MR) elastography for this non-invasive detection.

Material and methods

Animals: Groups of rats were imaged after being fed a standard diet (N=12), a choline-deficient diet up to 8 weeks to induce steatohepatitis (N=24) [1], a 2-week orotic acid diet to develop steatosis (N=8) [2] or were imaged 48 hours after injection of carbon tetrachloride to model acute liver injury (N=8).

MR elastography: The rats were placed under anesthesia in prone position on a piezoelectric based driver which generated mechanical waves at 300 Hz. Images were obtained on a 7 T Bruker Pharmascan animal imager. The propagation of the waves was imaged with a modified spin-echo sequence as described in [3]. The sinusoidal motion was sampled with four temporal steps. The full 3D displacement field was imaged inside the liver in the stationary wave regime and used to calculate the elasticity (dynamic modulus Gd) and the viscosity (loss modulus Gl) through a full inversion of the wave propagation problem [4]. Respiratory motion was triggered via a sensor monitoring the abdominal motion.

Histological, bio-chemical and bio-molecular analyses: Steatosis and fibrosis were quantified with morphometry and biochemical analysis. Myofibroblast activation was assessed with morphometry of alpha-smooth muscle actin. Expression of transforming growth factor β1, procollagen 1 and 3 as markers of fibrogenesis was evaluated with real-time reverse transcription polymerase chain reaction. Inflammation was scored at histology.

Statistical analysis: The comparison between groups was performed with the non-parametric Kruskall Wallis test, followed by two by two comparisons with the Wilcoxon rank-sum test. A Bonferroni correction was applied and a *P*-value ≤ 0.010 was regarded as statistically significant. The Spearman method was used for univariate correlations. Multivariate regression analysis was performed using the stepwise method.

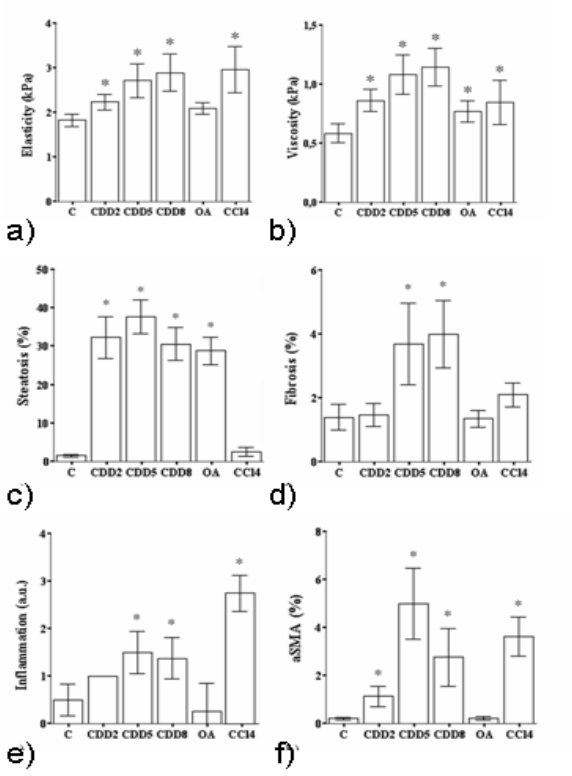

Figure 1: *Evolution of Gd (a), Gl (b), steatosis (c), fibrosis (d), score o inflammation (e) and stellate cell activation (αSMA) (f) according to th type of pathology. (* P ≤0.005 relative to control rats).*

Results and Discussion

In the rats with steatohepatitis, the elasticity and viscosity increased after two-week diet ($P < 0.005$ versus controls), while steatosis, inflammation myofibroblast activation, and increase of the other markers of fibrogenesis were observed. However, fibrosis appeared only after five weeks. In th rats with simple steatosis, the viscosity increased while elasticity remained constant. In the rats with acute liver injury, both elasticity and viscosit increased while fibrogenesis and inflammation were observed without substantial fibrosis or steatosis (Figure 1). Results of univariate analysis ar shown in Table 1. At multivariate analysis in all rats, the liver elasticity was only correlated to myofibroblast activation ($P < 0.001$, $r > 0.6$).

The results of this study in the rat suggest that in non-alcoholic fatty liver, MR elastography may be useful for the early detection of steatohepatitis b showing increased elasticity appearing before fibrosis development and linked to myofibroblast activation.

Table 1: *Correlations between visco-elasticity and percentages of fibrosis, steatosis, αSMA staining, expression of TGFβ1, type (I)- and (III) Procollagens and score of inflammation.*

	Fibrosis (%)	Steatosis (%)	Inflammation (a.u)	αSMA (%)	(I)-Procollagen	(III)-Procollagen	TGFβ1
Elasticity (kPa)	r = 0.61**	r = 0.33 (P = 0.018)	r = 0.53**	r = 0.67**	r = 0.70**	r = 0.64**	r = 0.57**
Viscosity (kPa)	r = 0.65**	r = 0.61**	r = 0.29 (P = 0.040)	r = 0.63**	r = 0.49**	r = 0.43**	r = 0.54**

** P ≤ 0.005

These results in rats also suggest that early diagnosis of non-alcoholic steatohepatitis in patients with fatty liver is feasible by observing increase hepatic elasticity at MR elastography, before the appearance of fibrosis. This might have important implications for the early detection and th treatment assessment of patients with non-alcoholic steatohepatitis.

References

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