

Optimal field strength for Molecular MR Imaging : detection of targeted Gd-based Contrast Agents on a Living Cell Monolayer

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INTRODUCTION

Great advances have recently been made in Molecular Magnetic Resonance Imaging (MRI), which allows non-invasive imaging of cellular or subcellular events. *In vivo* Molecular MRI has to cope with the inherently low sensitivity of MRI for the detection of exogenous Contrast Agent (CA), a small amount of molecular receptors and the complex biodistribution of the CA which influences its relaxation enhancement effects. High-molecular-weight CAs, such as nanoemulsions, which are able to carry several thousands of Gd³⁺ ions, are developed to overcome the low sensitivity of MRI. The r_1 relaxivity of such lipid-based paramagnetic CAs tends to decrease rapidly above 20-30 MHz, raising the question of their efficiency at high field. That effect, already studied *in vitro* [2], had not yet been highlighted in live cells.

The purpose of this study is to investigate experimentally the signal enhancement obtained with a paramagnetic nanoemulsion as a function of the field strength B_0 , taking into account of its Nuclear Magnetic Relaxation Dispersion (NMRD) profiles. The microfluidic set-up designed to follow-up the binding of a targeted CA on a *living cell* monolayer, developed at the lab [1], is used here to investigate the dynamic uptake of a Gd-based nanoemulsion by a cell monolayer at 2 static magnetic fields: 2.35 T and 4.7 T.

THEORY

A theoretical study by Girard et al [2] depicts the calculation of the Signal to Noise Ratio (SNR) and the Contrast to Noise Ratio (CNR) between tissues containing or not a CA, as a function of B_0 . It showed that the detection of commercial CA such as Gadolinium chelates was optimized at high magnetic fields. However, this does not apply to Gd chelates bound to macromolecular targets nor to high molecular weight CAs (such as dendrimers, liposomes or emulsions), which exhibit an increased relaxivity at low Larmor frequencies (due to an increase of the rotational correlation time).

The theoretical CNR between a free-CA tissue and the same tissue containing such CA reaches a maximum value at clinical magnetic fields (1.5T- 3T) and decreases above as displayed on figure 2. The NMRD r_1 profile of the Gd nanoemulsion used for our experiments has been fitted on experimental values and is shown in figure 1.

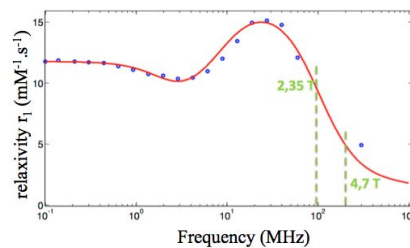


Fig.1: NMRD profile of the nanoemulsion used in this study

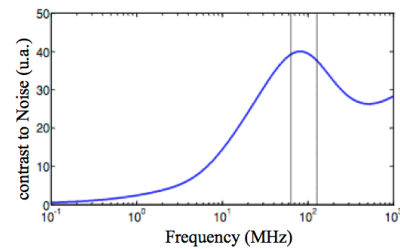


Fig.2: Theoretical CNR between a CA-free tissue and a tissue containing the nanoemulsion according to [2]

MATERIAL AND METHODS

Cells: A monolayer of HUVEC cells which overexpress the $\alpha_v\beta_3$ integrins was obtained by seeding 280 000 cells in the microfluidic channel (dimensions $0.4 \times 5 \times 50 \text{ mm}^3$) of a μ -Slide 10.4 Luer (Ibidi, Germany).

MRI: Experiments were carried out on both on a 2.35T scanner (Bruker, Germany) and a 4.7T scanner (Oxford). A 3D FLASH sequence was applied with $12.4 \mu\text{m}$ resolution perpendicular to the cell layer (cell layer's thickness $\approx 10 \mu\text{m}$), an in-plane resolution of $200 \times 400 \mu\text{m}^2$, $\text{TR}/\text{TE} = 75/3.7 \text{ ms}$. Unidimensional signal profiles were extracted by projection of the image matrix along planes parallel to the cell layer as previously described in [1].

Protocol: A flow of culture medium containing a $\alpha_v\beta_3$ -specific paramagnetic emulsion at a nanoparticle concentration of 3.75 nM was applied over the cell layer using a syringe pump and silicon tubing, with a velocity comparable to the blood's one in capillaries [3]. Every 15 minutes, the flow was stopped and the medium containing the nanoemulsion over the cell layer was flushed and replaced with CA free medium; an image was then acquired. The measurements were made with the same protocol for both magnetic fields.

RESULTS

SNR measurements were done on the medium without CA and were of about 7.5 and 21.1 at 2.35 T and 4.7 T respectively. Assuming an unloading sample (microfluidic channel), the expected SNR gain between these two fields would be of about 3.36. As previously described in [4], when the flow of nanoemulsion is applied over the cell layer, a part of the CA binds specifically to the $\alpha_v\beta_3$ integrins inducing a signal enhancement on the cell layer. The figure 3 compares the relative signal enhancements on the cell layer $G(t) = [S(t) - S(0)]/S(0)$, as a function of time, for the 2 magnetic fields. The mean signal enhancement measured between the 2 static fields is of about 2.7.

DISCUSSION & CONCLUSION

The sequences used in that protocol were optimized to increase the SNR on the medium without CA. The far more pronounced signal

enhancement of 2.7 at 2.35 T compared to 4.7 T can be explained by the decrease of the relaxivity r_1 (by a factor of 2) between those 2 fields. Our study tends to be in line with the theoretical study by Girard et al and demonstrates the lower efficiency of a paramagnetic nanoemulsion when the magnetic field increases, which raises the question of its use at high magnetic fields.

Ref: [1]. Gargam et al, ESMRMB 2011, [2]. Girard et al, ISMRM 2008, [3]. Stücker et al. Microvascular research, 1996, [4]. Gargam et al. ISMRM 2012

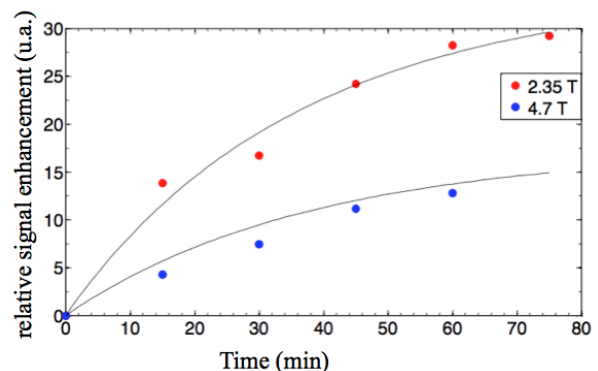


Fig. 3 : Relative signal enhancement on the cell layer following the application of a flow of targeted CA as a function of time.