Performance of hyperpolarized 1,4-$^{13}C_2$ fumarate in a murine lymphoma model *in vivo*, a new diagnostic agent for oncology

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Introduction: We have developed hyperpolarized 1,4-$^{13}C_2$ fumarate as a new promising metabolic marker for oncology. Metabolic parameters like up-take and conversion significantly influences imaging parameters like timing and concentration. Here we show a comparison between two substrates, hyperpolarized 1-^{13}C pyruvate, which previously has been shown to be an oncology marker[1,2] and the new agent, 1,4-$^{13}C_2$ fumarate. In a mouse EL-4 lymphoma model, the optimal timing and concentration are determined after which the SNR and contrast for the two substrates in the metabolic images are evaluated and compared.

Methods: The *in vivo* MR experiments were performed on a 2.35T Bruker Biospec Avance II system. EL4 tumor bearing c57BL/6 mice were anaesthetized and ECG, breathing rate and temperature was monitored (SA instruments). ^{13}C spectra were acquired with a 20-mm surface coil. Hyperpolarized 1,4-$^{13}C_2$-fumarate or 1-^{13}C pyruvate (2-50 mM) were injected *i.v.* (175 µl / 6 s) and either a slice selective FID or a double slice ^{13}C- CSI was acquired. 1,4-$^{13}C_2$-fumarate and or 1-^{13}C pyruvate was polarized to 30±3% or 26±3% respectively in the liquid state using a setup previously decribed[3].

Results and discussion: The optimal imaging timing was determined from the dynamic curves measured in a 20 mm slice going exclusively through the tumor (Figure 1). The substrates 1,4-$^{13}C_2$ fumarate and 1-^{13}C pyruvate both peak at the same time approximately 8 s after injection. The metabolites 1,4-$^{13}C_2$ malate and 1-^{13}C lactate on the other hand peak at 30 and 15 s respectively. The concentration was varied and the lowest concentration which gave full metabolite intensity and still a quantifiable substrate peak was determined. For 1,4-$^{13}C_2$ fumarate this was 10 mM. For 1-^{13}C pyruvate this was 20 mM due to a lacking substrate peak at lower concentrations. Therefore the SNR and contrast study was performed at 20 mM. For each substrate the optimal time was used to acquire the CSI. An example of the metabolic distribution of the two substrates and their metabolites in the same tumor is shown in Figure 2. The SNR for malate and lactate were determined in 6 animals and a total of 12 slices. The average SNR was 10.8 and 36.4 for 1,4-$^{13}C_2$ malate and 1-^{13}C lactate respectively. The contrast was determined as mean of the signal in voxels in-side and out-side the tumor. The average contrast was 7.2 and 4.9 for 1,4-$^{13}C_2$ malate and 1-^{13}C lactate respectively.

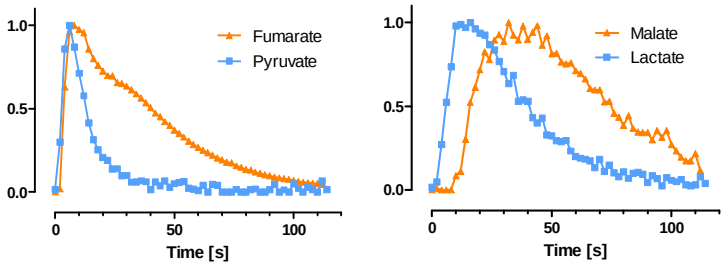

Fig 1: Dynamics for the substrate 1,4-$^{13}C_2$-fumarate and 1-^{13}C-pyruvate measured using a slice selective FID through the tumor in the mouse. All curves are normalized to the maximum signal of each individual substrate / metabolite.

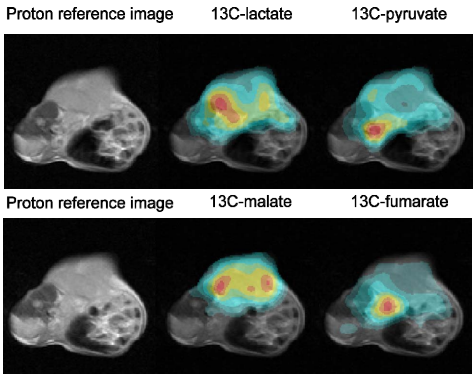

Figure 2: Metabolic distribution of 1-^{13}C-lactate/1-^{13}C-pyruvate and ^{13}C malate/1,4-^{13}C-fumarate in the EL4-lymphoma mouse model. The datase shown is representative for the contrast observed using the two substrates 1 ^{13}C-pyruvate and 1,4-^{13}C-fumarate.

Conclusion: The optimization of the timing for the new substrate fumarate proved very important. The conversion of 1,4-$^{13}C_2$-fumarate is slower than the one observed for 1-^{13}C-pyruvate and a difference in optimal timing of 15 sec was found between the two substrates. The concentration optimum was not so critical and a common compromise of 20 mM was chosen. In a comparative study the SNR on the metabolites 1,4-$^{13}C_2$ malate and 1-^{13}C lactate showed that 1,4-$^{13}C_2$ malate had 0.3 times the SNR of 1-^{13}C lactate. On the other hand the contrast for 1,4-$^{13}C_2$ malate was better than what was determined for lactate, 7.2 versus 4.9. This study showed that the conversion of 1,4-$^{13}C_2$ fumarate to 1,4-$^{13}C_2$ malate has comparable imaging qualities to the well known substrate/metabolite pair 1-^{13}C pyruvate/1-^{13}C lactate in this model.

Acknowledgement: GE Healthcare is gratefully acknowledged for financial support. K. Brindels group at Cambridge University for EL-4 protocol.

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