Quantification of Magnetically Labeled Cells with an SPIO Labeled Tumor Model in Rats

W. liu[1], E. K. Jordan[2], H. Dahnke[3], and J. A. Frank[2]

[1]Clinical Sites Research Program, Philips Research North America, Briarcliff Manor, NY, United States, [2]NeuroImaging Section, Laboratory of Diagnostic Radiology Research, Clinical Center, National Institutes of Health, Bethesda, MD, United States, [3]Philips Research Europe, Hamburg, Germany

INTRODUCTION

SPIO nanoparticles are used for cell labeling to monitor their migration and homing by MR imaging. Quantifying the number of labeled stem cells in target tissues in experimental models is of great importance to optimize dose and timing of cellular therapy in clinical trials. R2* (1/T2*) relaxation rate is a sensitive parameter for quantitative detection of intracellular SPIOs [1]. In this study, rats with implanted SPIO labeled C6 glioma tumors were imaged at different stages of the tumor development. The rapid proliferation of C6 glioma cells results in a variety of SPIO concentrations. Assuming SPIO nanoparticles stay within the tumor, the dilution of SPIOs corresponding to the tumor's growth can be estimated using the number of SPIO labeled cells divide by the tumor volume and should be linearly correlated with the mean tumor R2* value. This model provides a means to determine the quantitative relationship of labeled cells vs. R2* in an in vivo setting. Previous experiments have shown a linear correlation between tumor R2* and the number of labeled cells/mm^3 combining two different tumor cell lines in eight animals [2]. This longitudinal study allows for a concise analysis of the linear relationship over the course of individual tumor's growth and a cross validation of the correlation in different tumors.

METHODS

Animal Model: C6 glioma cells were labeled with ferumoxides-protamine sulfate (FEPro) complexes using procedures described previously [3]. Nude rats (n= 3) were implanted subcutaneously bilaterally with 1×10^6 FEPro labeled C6 glioma cells.

MRI: MRI was performed approximately in a two-week period after the inoculation of the tumor cells on a 3T Achieva whole-body scanner (Philips Medical System, Best, The Netherlands) using a dedicated 7cm rat solenoid rf-coil (Philips Research Laboratories, Hamburg, Germany). R2* maps were acquired with a multiple gradient echo sequence: TR/TE = 1540/16 ms, 13 echoes, 256 × 256 matrix, 17 slices, slice-thickness = 1.0 mm, FOV = 80 mm × 80 mm, NEX = 4.

Data Analysis: R2* relaxation rates were calculated by mono-exponential fitting using an in-house IDL software tool. The R2* relaxation of the tumor was calculated as the pixel wise average of R2* over the whole tumor volume. The number of labeled cells/mm^3 was determined as the number of implanted tumor cells divided by the tumor volume. The linear function for individual tumor was fitted from the data measured at different stages of each tumor. The detection limit was estimated as the slope corresponding to the R2* values that were the same as the maximum standard deviation of the linear function from each tumor. The gross linear correlation was fitted with all the data points from six tumors in three animals.

RESULTS

One rat was scanned three times and two rats were scanned twice during the experiment. Tumors varied in size from 750 mm^3 to 3683 mm^3 at the time of imaging, corresponding to the concentration of SPIOs approximately 542 to 2664 labeled cells /mm^3.

Figure 1 illustrates the R2* maps of an SPIO labeled tumor at different stages of the tumor development. Throughout the tumor growth, SPIO nanoparticles within the tumor were diluted and the corresponding R2* relaxation rates decreased as indicated by the increase in green coloring (from A to C). The individual linear correlation between average tumor R2* vs. number of labeled cells/mm^3, as well as the calculated detection limit from each tumor model, was summarized in Table 1. The average detection limit was estimated as 316 labeled cells/mm^3. The average R2* of the tumor demonstrated a very good gross linear correlation with the number of labeled cells/mm^3 (Figure 2), with a correlation coefficient of 0.90.

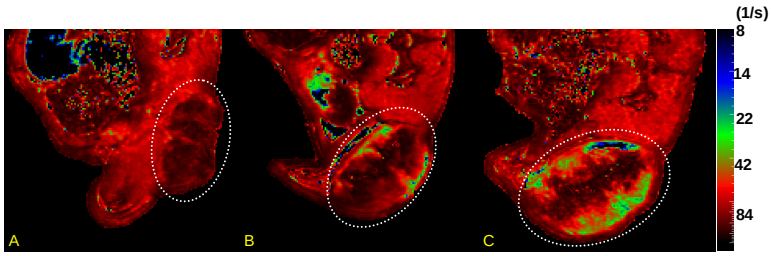

Figure 1. R2* maps of an SPIO labeled tumor at different stages. From A to C, the tumor grew bigger and tumor R2* decreased as indicated by the increase of green coloring**.** White circles indicate the tumor.

DISCUSSION

In this study, we investigated the quantitative relationship between the SPIO labeled cells and tissue R2* relaxation rate. Six tumors in three animals were evaluated at different stages of the tumor development. The individual linear relationship was consistent with the gross linear correlation from all the data set. Our data further demonstrate that R2* measurement is a reliable and sensitive tool for quantification of SPIO labeled cells. These results will allow quantitative assessment of magnetically labeled cells in vivo noninvasively.

Table 1. Individual linear relationship of R2* vs. number of SPIO labeled cells/mm^3 and the detection limit from each tumor.

Animal		Linear Function (y: R2*, x: cells/mm^3)	Detection Limit (cells/mm^3)
Rat No 1	Left	*y=0.0437x+20.51*	*321*
	Right	*y=0.0331x+27.40*	*262*
Rat No 2	Left	*y=0.0449x+3.59*	*328*
	Right	*y=0.0397x+3.24*	*443*
Rat No 3	Left	*y=0.0246x+22.09*	*254*
	Right	*y=0.0295x+23.10*	*646*
Average		*y=0.0304x+22.26*	*316*

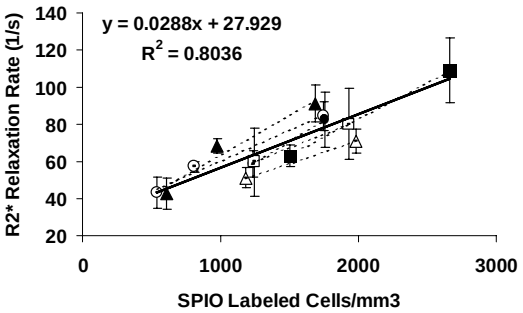

Figure 2. Tumor R2* vs. number of labeled cells/mm^3 in six tumors. Dotted lines indicate linear relationships from individual tumors. Solid line indicates the gross correlation fitted from all the data points. Data presented are the average R2* of the whole tumor volume. Error bars represent variations within each tumor.

Reference:

1. Yabloskiy DA. et al. MRM 1994;32:749-763;
2. Liu W. et al. ISMRM 2006 #680;
3. Arbab A. et al. Blood 2004;104(4):1217-1223.