Strategies for Improved Temporal and Spatial Resolution: In vivo Oxymetric Imaging using Time-Domain EPR

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Abstract

A radiofrequency (RF) time-domain electron paramagnetic resonance (EPR) instrumentation operating at 300, 600, 750 MHz for imaging tumor hypoxia with high spatial and temporal resolution is described. A high speed signal averager PCI board, with flexibility in input signal level and the number of digitized samples per free induction decay, is inducted in the receive arm of the spectrometer. This enabled effective and fast averaging of FIDs. Modification of phase encoding protocol and replacement of the GPIB-based handshake with a PCI-based D/A board for direct control of the gradient amplifiers decreased the gradient settling and communication overhead times by nearly two orders of magnitude. This is demonstrated by collecting a large number of 2D images successively and rapidly. These improvements show that it is feasible to achieve accurate, two dimensional pO_2 maps of tumor hypoxia with 1 mm^2 resolution with minimal artifacts, using a set of multiple gradient images within an acceptable measuring time of about 10 sec, and three dimensional maps in about 3 min.

Introduction

Low frequency electron paramagnetic resonance (EPR) has gone through considerable progress in the past 15 years (1). Several biomedical applications of EPR imaging are being pursued because of the unique capabilities in obtaining physiological information such as tissue pO2 quantitatively(2).The time-domain EPR techniques offer better temporal resolution as well as the Fellgett multi-channel sensitivity advantage for *in vivo* studies of small animals. We routinely employ the Single Point Imaging (SPI) method (3) of data acquisition in EPRI where the spatial resolution is not dependent on the spectral line width. However, the images can provide in vivo pO_2 by their pixel wise decay via T_2*. The image FOV for a given equally spaced phase-encoding steps depends on the delay in the single point acquisition and the images show a zooming effect proportional to the single point delay. The errors associated with the FOV scaling are avoided, by collecting several SPI data sets with multiple (at least three) G_{max} so that the same FOV and hence the image resolution is maintained. These data sets are re-assembled and re-normalized to obtain pixel-wise pO_2 with improved precision and significantly reduced artifacts. However, the image data requires a threefold increase in acquisition time. Therefore, *in vivo* imaging of tumor hypoxia at required accuracy and resolution, in acceptable measurement time, is challenging, and it demands further improvement in instrumentation in terms of enhancement of signal averaging by the incorporation of time-efficient data acquisition techniques, improvement in transmit and receive arms in terms of fast gradient switching and phase cycling of transmit pulse to reduce artifacts. Thus, all these challenges have been addressed in our RF FT EPR imager to achieve good spatial and temporal resolution in *in vivo* RF EPRI.

Modifications in the spectrometer design

(1) The original HP gradient amplifier was replaced by Kepco (Model BOP 20-20D) bipolar power amplifiers (Kepco Inc. Flushing, NY 11352). (2) A PCI analog output board CYDDA 04HRP (Cyber Research, CT 06405) plugged directly inside the host PC, provided 4-channels, 80 kHz and 16-bit D/A resolution. The 80 kHz speed leads to a fast conversion settling time of < 15 μs. (3) A zigzag raster system was used for the gradient ramping to get the k-space samples, to keep the overall gradient settling time to a minimum (Fig.1) . (4) Based on the T_1 of trityl radicals used as spin probe, we employed a TR of 5.5 μs with the corresponding Ernst flip angle of 80° for maximum steady state magnetization. The k-space matrix elements, collected in this way, were properly reshuffled during the post-collection data processing for Fourier reconstruction.

Results & discussion

With these modifications, the gradient settling time was brought to a fraction of a ms, and a two dimensional SPI data with 121 gradient steps (11 x 11 k-space) with four averages per gradient setting, gave an image with very little distortion in a total measurement time of just 2.6 ms (121 x 4 x 5.5 μs). 2D images with excellent temporal resolution(Fig.2), and 3D oxymetric imaging (3 images with different gradient steps within 5 minutes) could be carried out (Fig.3) with adequate spatial resolution (please see figure captions for details). With these improvements, we are able to generate 2D and 3D oxymetric EPR images in with very good spatial and temporal resolution.

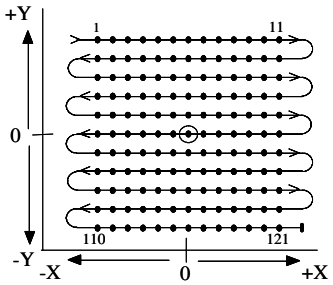

Fig.1 Two-dimensional zigzag raster corresponding to the collection of k-space points in the XY plane allowing only a gradual change in gradients with no large jump magnitude or large and abrupt change in sign. A similar approach is used for 3D k-space traversal as well.

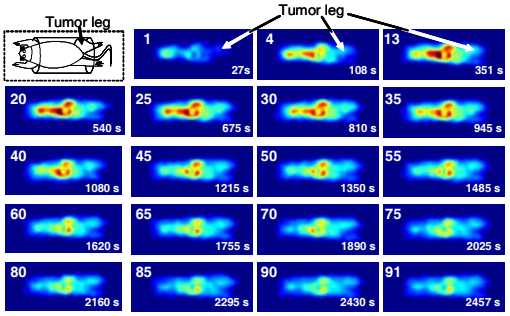

Fig.2 Sequential 2D pulsed EPR images (every 27s) of a C3H mouse, with SCC tumor on the top leg. The time in sec. after the injection of the spin probe is also given. (Image acquisition parameters: 15 x 31 gradient steps of 0.08 G/cm; pulse width 70 ns at 200 W nominal power; inter-pulse delay (TR) of 5.5 μs; FIDs summed per gradient step 10,000 at 500 Ms/s; Number of points sampled per FID 1600. Image was processed from a single point at 800 ns delay from the pulse).

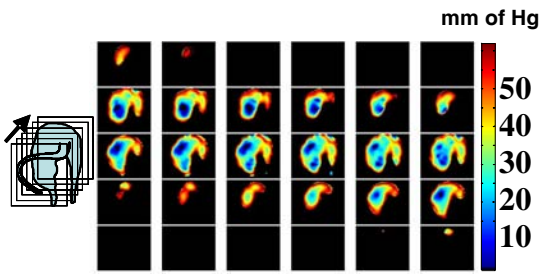

Fig.3 Coronal slices of pO_2 (alternate 0.6 mm slices) derived from a multi-gradient 3D SPI image that was carried out with 25 x 25 x 25 gradient steps and 0.8, 1.0 and 1.2 G/cm Maximum gradient. The pixel-wise slopes were used to determine the T_2* from which the concentration contribution were obtained by extrapolating the decay curve to "zero-time" immediately following the pulse. The greatly reduced pO2 in the tumor leg, the presence of the hypoxic cores, and the heterogeneity in the tumor oxygen distribution, etc. can be clearly seen.

References: (1) Berliner LJ, editor. *In vivo* EPR (ESR). Volume 18. New York: Kluwer Academic; 2003. (2) Elas M et al., *Magn Reson Med* 2003;**49**:682-691. (3) Matsumoto K et al., *Magn Reson Med* 2006;55:1157-1163.120