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EPR Image Based Oxygen Movies for Transient Hypoxia

Gage Redler^{a,b}, Boris Epel^{a,b}, and Howard J. Halpern^{a,b}

^aCenter for EPR Imaging In Vivo Physiology, Chicago, IL, USA

^bDepartment of Radiation Oncology, University of Chicago, Chicago, IL, USA

Abstract

Chronic hypoxia strongly affects the malignant state and resistance to therapy for tumours. Transient hypoxia has been hypothesized, but not proven to be more deleterious. Electron paramagnetic resonance imaging (EPRI) provides non-invasive, quantitative imaging of static pO₂ in vivo. Dynamic EPRI produces pO₂ movies, enabling non-invasive assessment of in vivo pO₂ changes, such as transient hypoxia. Recent developments have been made to enable Dynamic EPRI. Maximally spaced projection sequencing has been implemented to allow for more accurate and versatile acquisition of EPRI data when studying dynamic systems. Principal component analysis filtering has been employed to enhance SNR. Dynamic EPRI studies will provide temporally resolved oxygen movies necessary to perform in vivo studies of physiologically relevant pO₂ changes in tumours. These oxygen movies will allow for the localization/quantification of transient hypoxia and will therefore help to disentangle the relationship between chronic and transient hypoxia, in order to better understand their roles in therapeutic optimization and outcome.

1 Introduction

The important prognostic implications related to the oxygenation status of tumours have been known for decades¹. Chronic hypoxia, or chronically low oxygen concentration (pO₂), results in greater resistance to radiation therapy for cancer cells^{2, 3} and therefore has been correlated with radiotherapy treatment failure in humans⁴. Additionally, tumours exhibiting chronic hypoxia are more resistant to chemotherapy⁵, tend to grow more rapidly due to abnormal proliferation⁶, and have increased potential for metastatic progression⁷. These implications have led to a high level of interest in methods for measuring/imaging pO₂ deep in tissues.

Diffusion limited chronic hypoxia was the first form of hypoxia found in tumours⁸ and for many years was believed to be the only type of hypoxia present in tumours. However, more recent studies have found that perfusion limited transient hypoxia is present as well^{9–12} and may even be the major cause of hypoxia in tumours^{12, 13}. Studies suggest that transient hypoxia may be as important a determinant of cancer progression and patient prognosis as chronic hypoxia, if not more important. However, currently no definitive conclusions can be made due to inconsistencies in the data and methodological approaches from previous studies investigating the biologic effects of transient hypoxia versus those of chronic hypoxia. Furthermore, lack of data correlating a quantitative measure of transient hypoxia in vivo with treatment outcome currently precludes such a comparison¹⁴.

Electron paramagnetic resonance imaging (EPRI) has proven to be a robust imaging modality for studying tissue pO_2 in vivo. EPRI noninvasively acquires highly-resolved, both spatially ($\sim 1 \text{ mm}^3$ voxels) and in pO_2 (1–3 torr), 3D images of pO_2 in vivo^{15–19}. Acquisition time for our standard EPRI is on the order of 10 minutes, which is suitable for studying chronic hypoxia.

However, in order to use EPRI to study transient hypoxia, recent developments have been made to allow for improvements in temporal resolution (acquisition time on the order of 1 min) and accuracy. A novel acquisition method for maximally spaced projection sequencing has been implemented to allow for more accurate and versatile acquisition of EPRI data when studying dynamic systems (e.g., transient hypoxia). Principal component analysis filtering has been employed as well to enhance SNR and thus allow for improved temporal resolution. These improvements enable Dynamic EPRI, which can provide 4D EPRI based oxygen movies and allow for the necessary non-invasive studies investigating transient hypoxia to eventually help determine its biologic implications.

2 Methods

2.1 Maximally Spaced Projection Sequencing

EPRI is a projection based imaging modality with projections acquired in a spherical geometry. Therefore, the directions through which projections are acquired can be represented as points on the unit sphere (unit hemisphere sufficient due to radon transform symmetry). The influence of the uniformity of the spatial distributions of projections has been extensively studied^{20, 21}, but the uniformity with which the projections are acquired can still be optimized.

Before the MSPS method we used a polar azimuthal raster (PAR) method to acquire projections. The PAR method essentially rasters up one side of the unit hemisphere and then back down the other, which results in highly non-uniform distributions of projections throughout image acquisition. A new maximally spaced projection sequencing (MSPS) method has been implemented to achieve more uniform acquisition of projections. For the MSPS method, successive projections are acquired so that they are maximally spaced from projections that have already been acquired. In order to ensure uniformity for the final image, projection points are chosen from some final set of uniformly distributed points.

2.2 Principal Component Analysis Filtering

Principal component analysis is a method for defining a new orthonormal basis on which to express some given n -D data set. The basis vectors, or principal components (PCs), are chosen to be the eigenvectors of the covariance matrix for the data. These eigenvectors can be arranged in order of decreasing eigenvalues, which ensures that the first q PCs span a smaller q -D space and contain mostly correlated features, while the higher PCs contain mostly uncorrelated noise. Filtering of the data can be achieved with a low-order approximation by projection onto the first q PCs, thus selectively retaining relevant temporal patterns while discarding unwanted noise. PCA filtering is applied to dynamic EPRI studies as a pre-reconstruction spatiotemporal filter for projection data²².

3 Results

3.1 Maximally Spaced Projection Sequencing

The distribution of projections at intermediate steps throughout imaging converge to uniformity more rapidly for the MSPS method as successive projections are acquired than for the PAR method. This yields intermediate images reconstructed from incomplete projection sets that approximate the final image more closely and earlier during imaging (Fig. 1).

Acquisition using the MSPS method affords the versatility to adjust temporal resolution to best fit the experiment and data following acquisition. This can be done simply by reconstructing temporally adjacent intermediate subsets of projections. This can allow for the visualization of important physiologic temporal changes that may not have been resolved with the temporal resolution of the original study (Fig. 2).

3.2 Principal Component Analysis Filtering

Simulation studies show that PCA filtering can significantly enhance SNR (by a factor of 3.8 ± 0.6) and allow for the order of magnitude improvement in temporal resolution necessary to obtain EPRI oxygen movies with 1 minute frames. PCA also helps to emphasize relevant temporal features. PCA is suited for data with large portions having common features and a small number of such features. Studies suggest that this is the case for tumors exhibiting transient hypoxia²². PCA filtering of Dynamic EPRI data is demonstrated in an experiment with a mouse breathing air (21% O₂) followed by carbogen (95% O₂) and then air again. One minute frames from this experiment are displayed and the tissue response to breathing gas changes can be clearly resolved.

4 Discussion and Conclusions

Important tools such as the MSPS acquisition method and PCA filtering have been developed for EPRI in order to enable Dynamic EPRI with temporal resolution on the order of 1 minute. The enhancements and versatility provided from these techniques enable 4D EPRI based oxygen movies well resolved spatially, temporally, and in pO₂. Dynamic EPRI therefore is an ideal modality to study physiologically relevant temporal pO₂ changes in vivo without perturbing the system of interest. These oxygen movies will allow for the spatial localization as well as quantification (frequency, amplitude, etc.) of transient hypoxia in vivo and therefore enable the eventual determination of its clinical relevance.

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Fig. 1. Uniformly distributed projections throughout imaging for the MSPS method results in intermediate images that approximate the final image more rapidly than for the PAR method.

Fig. 2.
(a) Single MSPS image acquired over 36 minutes with mouse breathing air for first 18 minutes followed by carbogen for second 18 minutes. (b) Two 18 minute intermediate images that are subdivisions of the full image that now allow resolution of tissue response to increased breathing gas oxygen (tumor outlined in red).

Fig. 3.

One minute frames from a PCA filtered Dynamic EPRI oxygen movie. Corresponding changes in breathing gas shown with red line. The mouse starts off breathing air and is switched to carbogen and then back to air. The temporal changes in tissue (tumor outlined in red) pO₂ are clearly resolved with such EPRI based oxygen movies.