Quantitative T1 and T2* assessment of VX2 tumour oxygenation in response to hyperoxia and hypercapnia: comparison with invasive measures and DCE-MRI

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Introduction: Hypoxic tumour regions have a reduced response to radiotherapy and may require 2-3 times the radiation dose to achieve the same level of cell death. Strategies to increase tumour oxygenation is a major area of ongoing research, and typically involve altering the patient's fraction of inspired O2 and/or CO2 (FiO2 and/or FiCO2) alone or in combination with pharmacological agents. Incorporation of non-invasive imaging of tumour oxygenation could enable subject-specific delivery of inspired gas concentrations to maximize treatment efficacy. Numerous studies employed T1 and T2* relaxation times to track tissue O2 levels in normal tissue1 and tumours2; however, simultaneous blood flow and volume changes during inspired gas modulation obscure the association between MR relaxation times and tissue partial pressure of O2 (pO2). This is further complicated in tumours due to the abnormal vasculature, variable baseline relaxation times and dependence on the host tissue response. Beyond relaxation time measurements, dynamic contrast enhanced (DCE) MRI has also shown promise as a predictor of tumour sensitivity to radiotherapy, based on the relationship between vascular characteristics and tumour hypoxia. The purpose of this study was to characterize T1 and T2* responses to 100% O2, and carbon monoxide with three different CO2 concentrations in a VX2 tumour model and compare the responses with quantitative DCE parameters, fibre optic measurements, and microsphere measures.

Methods: VX2 cell suspensions were injected into the thigh muscle of six New Zealand white rabbits to induce growth of a soft tissue carcinoma. MRI scans were performed 12 - 23 days following VX2 cell implantation to provide a range of tumour development. One day following the MR study, invasive tissue oxygenation and tissue perfusion measurements were obtained. Five gas challenges were delivered during MR imaging and invasive studies: i. air (21% O2, balanced N2); ii. 100% O2; iii. 3% CO2 and 97% O2; iv. 6% CO2 and 94% O2; and, v. 9% CO2 and 91% O2. MR acquisitions were performed on a 1.5 T GE Signa EXCITE MRI, using a 3° receive-only surface coil positioned under the rabbit's thigh muscle for imaging the tumour. A separate transmit-receive knee coil for acquisition of the arterial input function (AIF) from the aorta. Quantitative T1 values were generated for each gas using 3D FSPGR scans with three different flip angles (FA1). TR = 6.55 ms, TE = 2.7 ms, FA = 90°, NEX = 5. Five gas challenges were delivered during DCE-MRI data acquisition that captured the aorta in one slice. Imaging parameters included: TE = 1.37 ms TR = 3.01 ms, FA = 15°, FOV = 120 mm, matrix = 128 x 96, SL = 3 mm, number of slices (Nsl) = 10, NEX = 4. Quantitative T2* measurements were collected for each gas with a 2D multi-echo GRE sequence: TR = 10 ms, 6 equi-spaced TEs = [21.47, 1] ms, FA = 30°, FOV = 120 mm, matrix = 192 x 160, SL = 3 mm, Nsl = 6, and NEX = 3.

DCE-MRI data were collected using a dual-bolus approach. The AIF was collected in the knee coil using an initial prebolus (0.04 mmol/kg) Gd-DTPA (Magnevist, Berlex Canada) with a sagittal TRICKS acquisition that captured the aorta in one slice. Imaging parameters included: TE = 1.37 ms TR = 3.01 ms, FA = 20°, FOV = 190 x 90, Nsl = 10, SL = 3 mm, NEX = 0.75, and temporal resolution = 0.554 s. After 15 – 25 minutes, quantitative T1 values were collected in the tumour using the 3° surface coil. Full bolus (0.16 mmol/kg) DCE-MRI scans were then obtained using a 3D FSPGR scan with the following imaging parameters: TE = 2 ms, TR = 5.12 ms, FA = 15°, FOV = 120 mm, matrix = 128 x 96, SL = 3 mm, Nsl = 10, NEX = 0.75 and temporal resolution = 2.925 s.

Time processing was performed offline in Matlab. T1 maps were calculated using the SPGR steady state signal equation with flip angle correction1. T2* maps were calculated using a monoexponential signal decay3. Regions-of-interest (ROI) were outlined on T1 and T2* maps in the: 1. tumour rim, 2. tumour core, 3. necrotic tumour region, and 4. normal muscle to generate mean T1 and T2* values for each region. For DCE quantification, the AIF was first extracted from an ROI in the aorta from the prebolus scan and converted to concentration using the linear relationship between ΔR1 and contrast concentration. Tissue uptake curves were extracted from the four ROIs described above and converted to contrast agent concentration, using the measured T1 values. The tissue contrast uptake curve was modeled using the adiabatic approximation tissue homogeneity model (AATHM)4 to quantify the contrast extraction fraction (E), plasma flow (Fp), mean capillary transit time (Tc), and interstitial volume fraction (vI)

Tissue pO2 measurements were performed using the OxyLite system (Oxford Optronics, Oxford, UK), which utilizes fluorescence lifetime measurements to quantify absolute tissue pO2. Two probes were inserted directly into one of the four regions isolated in MR imaging analysis (e.g., tumour periphery). Invasive perfusion measurements were performed using 15 um diameter neutron-activated microspheres.

Results: Figure 1 provides example DCE MRI images at peak contrast, with the corresponding T1 and T2* maps from two different rabbits. ΔT1 and ΔT2* values relative to air breathing (21% O2, balanced N2) are reported in Table 2. We observed a consistent ΔT1 decrease and ΔT2* increase across all four gas challenges in all tissue regions. Effects of increasing carbon monoxide CO2 concentration on ΔT1 and ΔT2* are shown in Figure 3, where ΔT1 values are consistently reduced, with a significant negative correlation was observed between ΔT1 and ΔPaCO2 in the core tumour region (r = -0.54, p < 0.05), while the vI was still variable and did not follow a trend. For the DCE-MRI data, the only significant relationship existed between Fp and ΔT1 (r = 0.58, p < 0.05). Oxylite invasive measurements of pO2 revealed a substantial increase at the transition to 100% O2 that was sustained for the duration of the experiment. Microsphere perfusion values show increased perfusion during 100% O2 with a small decrease after administration of 3% CO2 in normal and necrotic regions.

Discussion: In this study, all hypoxic gas challenges substantially increased tumour pO2 levels, which was reflected in decreased T1 and increased T2* measurements, likely due to higher dissolved oxygen and higher venous HBO2-to-Hb ratio, respectively. A major observation was that in the VX2 tumour model, CO2 effects in carbon did not influence MR relaxation times compared with 100% O2, which suggests similar tumour oxygenation levels may be achieved with or without a CO2 component. This observation is supported by previous work that showed independence of pO2 levels with respect to CO2 concentration in inhaled carbon dioxide mixtures3. Results of the current study further support the use of MR relaxation times to aid adaptive strategies for improving tumour pO2 for radiotherapy.