Acta Oncologica, 2015; 54: 1521–1528

Taylor & Francis
Taylor & Francis Group

ORIGINAL ARTICLE

Early changes in perfusion of glioblastoma during radio- and chemotherapy evaluated by T1-dynamic contrast enhanced magnetic resonance imaging

SØREN MØLLER[1], MICHAEL LUNDEMANN[1], IAN LAW[2], HANS S. POULSEN[1,3], HENRIK B. W. LARSSON[2,4] & SVEND AAGE ENGELHOLM[1]

[1]*Department of Oncology, Section for Radiotherapy, Rigshospitalet, University of Copenhagen, Denmark,*
[2]*Department of Clinical Physiology, Nuclear Medicine & PET, Rigshospitalet, University of Copenhagen, Denmark,*
[3]*Department of Radiation Biology, Rigshospitalet, University of Copenhagen and*
[4]*Functional Imaging Unit, Department of Clinical Physiology, Nuclear Medicine & PET, Rigshospitalet, University of Copenhagen, Denmark*

ABSTRACT

Background. The survival times of patients with glioblastoma differ widely and biomarkers that would enable individualized treatment are needed. The objective of this study was to measure changes in the vascular physiology of tumor using T1-dynamic contrast enhanced magnetic resonance imaging (DCE-MRI) in patients with glioblastoma during early stages of radio- and chemotherapy (Tx) and explore possible correlations with treatment outcomes.

Material and methods. An exploratory prospective study was planned. Patients underwent DCE-MRI at baseline, after approximately one and six weeks of Tx and three and six months post-Tx. DCE-MRI at three Tesla generated maps of blood flow (BF), blood volume (BV), permeability (Ki) and volume of distribution (Vd) using a combination of model-free deconvolution and Patlak plots. Regions of interest in contrast enhancing tumor and in normal appearing white matter were contoured. Progression-free survival (PFS) was the primary clinical outcome. Patients with PFS >6 months were compared with those with PFS <6 months. Parameters of vascular physiology and changes in these during Tx were compared for these two groups at all time points using non-parametric statistics.

Results. Eleven eligible patients were included and 46 DCE-MRI examinations were carried out. BF in tumor increased for all patients early during Tx ($p = 0.005$) and then fell to a level below baseline at post-Tx examinations ($p = 0.016$). A similar but non-significant trend was seen for tumor BV. There was no detectable difference between patients with PFS >6 months versus PFS <6 months with regards to baseline values or changes during and after Tx.

Conclusions. Although no correlations to outcomes were found, the results of this exploratory study may be hypothesis generating and will be examined in a larger patient group.

Glioblastoma is the most common type of primary brain cancer and among the most devastating of all cancers. The standard treatment consists of surgery followed by combined chemo-radiotherapy [1]. However, this treatment regimen is lengthy (+8 months) and many patients deteriorate due to disease progression before completing it. Individual treatment options would be preferable but biomarkers for selecting the best treatment on an individual basis are needed.

Imaging biomarkers may include metabolic information such as tumor uptake of radiolabeled molecules using positron emission tomography (PET) or parameters of vascular physiology, such as tumor blood volume (BV) using computer tomography (CT) or magnetic resonance imaging (MRI). The most commonly used method for evaluating brain tumor vascular physiology is the dynamic susceptibility T2*-weighted (DSC) MRI method. It has proven useful for predicting glioma grade before surgery [2] and is FDA-approved and widely commercially available. Disadvantages include susceptibility to artifacts and the need for pre-bolus injection of contrast agent to correct for leakage [3]. Most

Correspondence: S. Møller, Engbakkevej 12, 2920 Charlottenlund, Denmark. Tel: +45 27141517. E-mail: dr.smoller@gmail.com.

(Received 14 May 2015; accepted 10 June 2015)

ISSN 0284-186X print/ISSN 1651-226X online © 2015 Informa Healthcare
DOI: 10.3109/0284186X.2015.1063777

previous studies evaluating brain tumor vascular physiology have focused on tumor BV [4,5]. We have previously described a method of generating cerebral blood flow (CBF) and cerebral blood volume (CBV) maps and permeability maps (CBKi) by dynamic contrast enhanced (DCE)-MRI at 3.0 T using a single bolus injection of paramagnetic contrast agent [6,7]. Leaky blood-brain barrier (BBB) is recognized as an important hallmark of malignancy in glioma but only a few studies have attempted to analyze BBB Ki in gliomas undergoing treatment in relation to outcomes [8,9].

An exploratory prospective study of patients receiving standard concomitant chemo-radiotherapy for glioblastoma was carried out. Patients were subjected to DCE-MRI scans at baseline, after approximately one and six weeks of radiotherapy (RT) and three and six months post-RT (MRI_1-MRI_5). The aim was to describe possible changes in tumor vascular physiology [BF, BV, Ki and blood volume of distribution (Vd)] using this technique and to explore possible correlations to clinical outcomes.

Material and methods

Patients with glioblastoma WHO grade IV referred to concomitant chemo-radiotherapy [1] were eligible to be included. The study was approved by the local Ethics Board (case number: H-D-2008-002, date of acceptance: 11 January 2012) and carried out in accordance with the Helsinki Declaration. Written informed consent was required. The study was observational and results achieved with DCE-MRI were not known to the treating physicians at the time of treatment. Patients went off-study at either disease progression, completion of the last DCE-MRI scan or withdrawal of consent. The time of progression was determined using the patient charts and decisions made in clinical practice in each individual case. These were based on the Macdonald criteria.

Patients

Inclusion criteria were: histologically confirmed glioblastoma, age 18–70, performance status 0–2, signed informed consent. Measurable residual tumor on post-operative MRI was mandatory, but as the study was exploratory there was no prespecified lower limit to tumor volume. Exclusion criteria were: contraindications to MRI or contrast injection, such as pacemaker, non-compatible metallic implants, reduced kidney function (defined as glomerular filtration rate < 60 ml/min), previous allergic reaction to MRI contrast agent, pregnancy and claustrophobia.

Radiotherapy

Target definition for RT was based on MRI and ^{18}F-fluoro-ethyl tyrosine PET (^{18}F-FET PET). ^{18}F-FET PET has been used in the standard treatment setting at our department since 2011. The gross tumor volume (GTV) consisted of a tumor volume defined by MRI (GTV MRI) and a tumor volume defined by ^{18}F-FET PET. The GTV MRI was defined by a radiologist using the contrast enhanced MRI sequence of a planning scan performed at RT planning (Siemens Magnetom Espree 1.5 T). Surgical cavities were contoured as part of the GTV MRI.

The total target volume consisted of the united volume of GTV MRI and GTV PET. A 20-mm margin was added to this volume to form the clinical target volume (CTV), to which 2 mm was added to form the planning target volume (PTV). A total dose of 60 Gray (Gy) was delivered in daily 2-Gy fractions over six weeks to the PTV. RT was delivered using volumetric modulated arc therapy (VMAT) on a Novalis Tx accelerator (Varian/BrainLab). Oral temozolomide was administered at 75 mg/m^2/day during RT and for six cycles (5 days/28 days) at 200 mg/m^2/day following RT [1].

DCE-MRI

DCE-MRI was performed on a 3.0 T MRI unit (Achieva; Philips Healthcare). Scans were acquired at baseline (0–7 days before start of RT [MRI_1]), following 10 Gy of RT (MRI_2), following 50 Gy of RT (MRI_3), three months post-RT (MRI_4) and six months post-RT (MRI_5).

DCE imaging used a saturation-recovery gradient–echo sequence with 120 ms delay (TD) between prepulse and the first readout pulse. A 90° non-selective saturation prepulse covering the head and thorax was applied before each sequence. Readout pulses had a flip angle of 30°, TR = 3.9 ms, TE = 1.9 ms, centric phase ordering, parallel imaging factor 2, acquired matrix 96×61 interpolated to 256×256, field-of-view 230×182 mm^2. Five slices with a thickness of 8 mm were generated. The most caudal slice was placed at the level of the internal carotid artery for determination of the arterial input function (AIF). The remaining four slices (interslice gap = 1.5 mm) were placed at the level of the tumor [6]. In case of tumors that were too large to be covered by four slices, the procedure was repeated in a second run immediately following the first. A separate T1 measurement was acquired before the first run and also before the second run, if two runs were performed [10].

The bolus of contrast (Dotarem 0.1 ml/kg body) was injected using an automatic injector and was followed by 20 ml saline. The bolus tracking had a time

resolution of 1.25 s for 250 time points in approximately 6 min.

The T1 measurement was generated before bolus injection by varying TD from 0.12–10.0 s, creating T1 and M0 maps corresponding to the dynamic images obtained during the bolus passage. The AIF was generated by placing the most caudal slice at the level of the vertical segment of the internal carotid artery. The larger of the two internal carotid arteries was contoured and the pixel within this area with the largest signal change at bolus passage was used for subsequent calculations. To minimize the partial volume effect due to the small diameter of the vessel, the AIF was scaled to the venous output function derived from the sagittal sinus [11]. BF was estimated by model-free deconvolution (Tikhonovs method) as previously described by Larsson et al. [6]. The general mathematical expression for this calculation is:

$$C_t(t) = f\, c_a(t) \otimes r(t)$$

where $C_t(t)$ = tissue concentration at time (t), $C_a(t)$ = arterial concentration at time (t) (AIF), f = flow and r(t) = impulse residue response function.

The BV and Ki was estimated by a graphical evaluation ('Patlak plot') of the following equation using assumptions of unidirectional flux of a low-permeating substance in a two-compartment system [12]:

$$\frac{C(a)}{C(t)} = Ki\frac{\int_0^1 C_a(t)dt}{C_a(t)} + BV$$

It is apparent that Ki is the slope of the fitted line, whereas BV is the intersection at t = 0. An example is provided in Figure 1. In-house software for MATLAB R2012a (MathWorks®, Inc.) was used for these calculations.

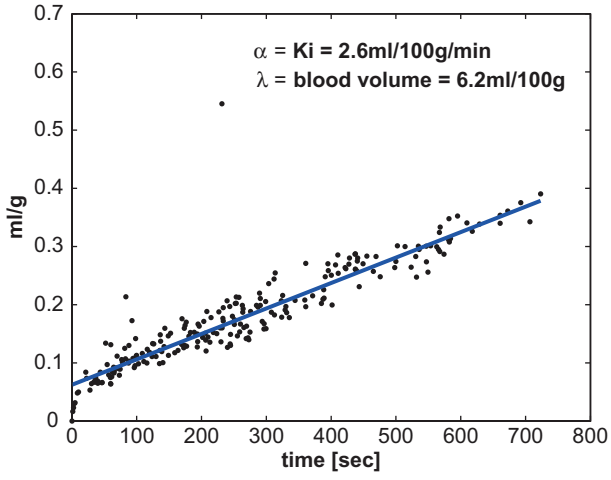

Figure 1. Example of a Patlak plot derived from an area within the tumor ROI of a patient. The regional permeability (Ki) in this area is estimated as the slope of the curve during steady-state, whereas the blood volume is derived from the intercept of the Y-axis as described in the section *DCE-MRI*.

In addition to the DCE sequence acquisition described above, the following sequences were also generated: T1W-3D, T2W, fluid attenuation inversion recovery (FLAIR), diffusion tensor imaging (DTI), blood oxygen level BOLD (in resting state) and arterial spin labeling (ASL).

Regions of interest and registration of images

Regions of interest (ROIs) were contoured on MRI that was registered to a planning CT scan. Two three-dimensional 3D ROIs were defined for measurements of vascular physiology: 1) tumor volume defined by contrast enhancing tumor on a T1 + Gd sequence used for RT planning. This volume was modified to exclude resection cavities; 2) normal appearing white matter (NAWM) drawn manually in healthy appearing white matter in the contralateral hemisphere within the section of brain encompassed by the DCE slices. The ROIs remained unchanged throughout the study.

For each of the five MRI examinations (MRI_1-MRI_5), the CT scan with the ROIs was rigidly registered to the anatomical T1W-3D sequence (isotropic, $1mm^3$) and each T1 sequence was subsequently registered to the axial T2W slices. Registrations were performed with the Functional Magnetic Resonance Imaging of the Brain Analysis (FMRIB) Software Library using either mutual information or normalized mutual information [13]. Each registration step was visually verified and accepted if the overlap of contour to tumor was deemed greater that 90%. The two registrations were combined and used to transform the CT-based ROIs to the DCE-slices.

Normalization to normal appearing white matter (NAWM)

All parameters of tumor vascular physiology [BF, BV, Ki and blood Vd] were normalized to corresponding values within NAWM for each run.

Statistics

Baseline vascular physiology values in ROIs were calculated as mean values for NAWM and median values for tumor ROIs. The patients were dichotomized according to their progression-free survival (PFS) (longer or shorter than six months from the date of diagnosis). Statistical comparison between these groups was carried out using the Mann-Whitney test whereas paired data were compared using the Wilcoxon test. A univariate Cox regression analysis was carried out to test the prognostic value of changes

in vascular physiology values compared to baseline. p-Values < 0.05 were considered significant. All statistical analyses were carried out using SPSS 19 (IBM Corp.).

Results

Eleven patients were included and they underwent a total of 46 scans. All patients completed RT and received daily temozolomide during the concomitant stage of treatment as prescribed. The number of patients scanned at each time point were: MRI_1 $n = 11$; MRI_2 $n = 10$; MRI_3 $n = 11$; MRI_4 $n = 10$; MRI_5 $n = 4$. At the time of MRI_5, (six months after RT), six patients had experienced progressive disease and one patient had withdrawn consent. The median PFS was 7.8 months. We identified one case of pseudo-progression [14], where the contrast enhancing lesion grew significantly at the first post-RT evaluation (MRI_4), but subsequently stabilized. Two patients currently remain free from disease progression + 3 years from time of diagnosis.

Figure 1 is an example of a Patlak plot, from which the tumor BV and Ki are quantified. This example was derived from an area within the tumor ROI in the patient with the highest measured tumor Ki (median Ki in tumor = 3.6 ml/100g/min). Similar plots were generated in other cases, verifying the linearity of the model. We have previously shown by use of simulation that for this set-up, the assumptions related to the Patlak model are satisfied for Ki up to around 3 ml/100g/min. If higher Ki is encountered, the use of two-compartment models is often recommended [7].

Figure 2 shows an example of the definition of ROIs (tumor and NAWM). As this study was exploratory, there was no prespecified lower limit to the tumor volume for eligibility to be included, but measurable disease as defined by the Macdonald criteria was mandatory. The median volume of tumor ROI and of NAWM was 34.3 cm^3 (range 11.3–89.7 cm^3) and 16.7 cm^3 (range 1.7–28.6 cm^3), respectively. To examine the possibility of partial volume effects, the correlation between tumor volume and parameters of vascular physiology was evaluated using Spearman's rank correlation coefficient, r_s. No statistically significant correlations could be identified (r_s was -0.23, -0.40, -0.51, -0.37 for BF, BV, Ki and Vd, respectively, using absolute, non-normalized values), but the smallest tumor volume in the study (measuring 11.3 cm^3) was found to also have the lowest values of vascular physiology parameters. NAWM ROI's were generally smaller but no correlations were found here either (r_s was -0.30, -0.36, -0.56, -0.44 for BF, BV, Ki and Vd, respectively).

At baseline, all vascular physiology values (BF, BV, Ki and blood Vd) were higher in tumor than in NAWM in all cases except one (where blood $flow_{NAWM} >$ blood $flow_{tumor}$). Please refer to Table I, which shows absolute values of vascular physiology at baseline. Measurements of vascular physiology in NAWM at baseline were compared with all subsequent examinations. Generally, these parameters did not change significantly within the NAWM. However, a significant difference was found between NAWM Ki at MRI_1 and MRI_4 (mean: 0.2 ± 1 vs. 0.1 ± 0.1; $p = 0.02$). This was not significant at MRI_5.

Figure 3 shows changes in tumor BF, BV and Ki (normalized to NAWM) over time for all patients and divided into patients with PFS > 6 months ($n = 6$) versus patients with PFS < 6 months ($n = 5$). There was a significant increase in tumor BF at MRI_2 compared to MRI_1 which applied to all patients.

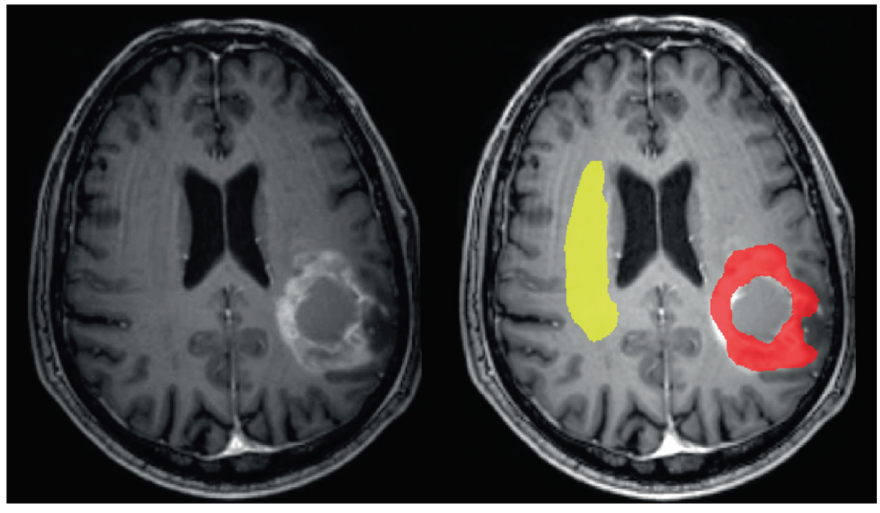

Figure 2. Example showing delineation of regions of interest on T1w-contrast enhanced MRI. Yellow mask on rightmost image: normal appearing white matter. Red mask: tumor. Note that the central tumor cavity was not included in the ROI.

Table I. Baseline values of parameters of vascular physiology for NAWM and tumor (mean ± standard deviation).

		NAWM (n = 11)	Tumor (= 11)	p-Value (Tumor vs. NAWM)
Blood flow	ml/(100g/min)	11.6 ± 3.4	18.9 ± 7.7	0.006
Blood volume	ml/100g	1.0 ± 0.4	4.2 ± 3.2	0.003
Permeability (Ki)	ml/(100g/min)	0.2 ± 0.1	1.2 ± 0.9	0.003
Volume of distribution	ml/100g	1.7 ± 0.8	7.5 ± 5.2	0.003

NAWM, normal appearing white matter.

Tumor BF decreased again at MRI_3 and MRI_4. At MRI_4, values of tumor BF and tumor BV were significantly lower than at baseline for the whole group, but there was no significant difference between patients with PFS > 6 months versus PFS < 6 months. Comparing the absolute, non-normalized, values (not shown) of tumor BF and tumor BV showed a similar statistically significant decrease from MRI_1 to MRI_4, however, the absolute increase from MRI_1 to MRI_2 did not prove statistically significant.

Using univariate regression analysis, the hazard ratios for progression and death for changes from scan 1 to 2 and from scan 2 to 3 were analyzed. No statistically significant correlations with outcomes were seen.

Figure 4 shows perfusion parameter maps at four time points for a patient with a short PFS of approximately four months. At the fourth time point, the patient deteriorated clinically and underwent re-operation. The pathology report confirmed the diagnosis of tumor progression.

Discussion

In this exploratory prospective study of glioblastoma patients receiving standard radio-chemotherapy, we found that tumor BF determined by DCE-MRI was significantly increased one week into RT compared with baseline. This has not to our knowledge been described previously in the literature, but Cao et al. have described a similar increase in tumor BV during early stages of RT [4] as well as an increased Ki [9]. In studies of human tumor xenografts, an early stress response to ionizing radiation has been demonstrated within 24 hours of exposure consisting of a dose-dependent increase in vascular endothelial growth factor (VEGF) expression [15]. VEGF promotes formation of new blood vessels [16] and blockade of the

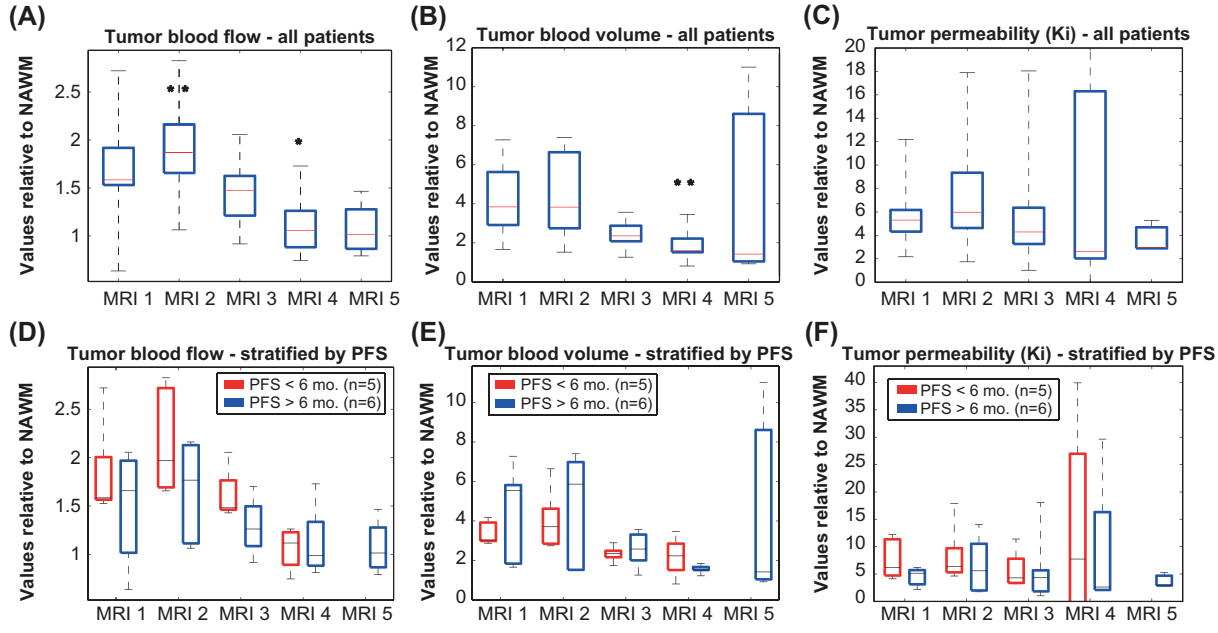

Figure 3. Values of vascular physiology parameters normalized to normal appearing white matter (NAWM) at all time points. (A–C) all patients (D–F) stratified by length of progression-free survival (shorter vs. longer than 6 months). Thin line denotes the median. Boxes include 25–75% of values. Whiskers include all data points. $N_{MRI\ 1} = 11$, $N_{MRI\ 2} = 10$, $N_{MRI\ 3} = 11$, $N_{MRI\ 4} = 10$, $N_{MRI\ 5} = 4$. *Significant change from MRI 1 (p-value < 0.05); ** Significant change from MRI 1 (p-value < 0.005).

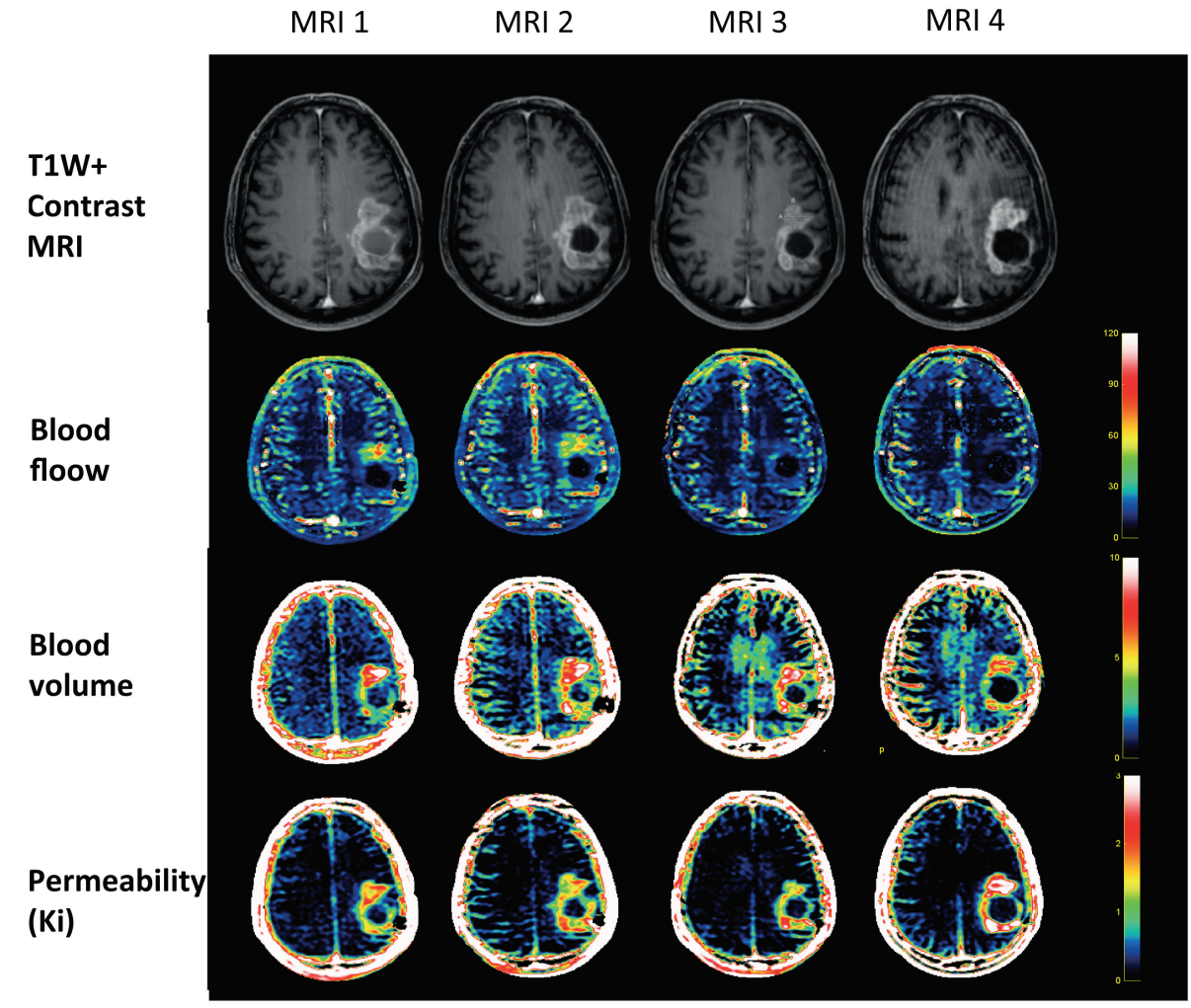

Figure 4. Vascular physiology parameter maps for a patient with a short progression-free survival of approximately 4 months at time points MRI_1 to MRI_4. The patient had disease progression at the last time point, but only the permeability (Ki) map reflects this whereas both tumor blood flow and tumor blood volume are lower than at baseline. Disease progression was confirmed histologically following re-resection. The color keys on the right show corresponding absolute values of blood flow [ml/(100g/min)], blood volume (ml/100g) and Ki [ml/(100g/min)].

VEGF pathway has early and measurable effects (~24 hours) on vascular physiology parameters (BV and Ki) [17]. It is therefore also possible that the early increase in BF was due to a transient rise in VEGF that later fell due to treatment response. However, this is a theoretical hypothesis and not – to our knowledge – supported by clinical evidence. Cao et al. proposed that corticosteroid treatment during RT could influence BF but we do not find this likely to be important in our study, as there was no systematic change in treatment or dosage of steroids. Due to the lack of an untreated control group in our study, we cannot rule out that early increases in BF and BV are a physiological phenomenon related to the regrowth of tumor in the weeks following surgery, rather than an effect of RT.

The early increase in tumor BF was analyzed in relation to outcomes but we could detect no differences between patients with PFS $>$ 6 months versus PFS $<$ 6 months. At MRI_3 and MRI_4, tumor BF was markedly decreased but still no correlations to outcomes were seen, which was also evident from the parameter maps shown for a patient in Figure 4. Taken together, it would appear that tumor BF is alterable by chemo-radiotherapy but not an important marker of tumor malignancy. Similarly, a leaky BBB is effectively sealed by the anti-VEGF antibody bevacizumab, but treatment with this drug has not been shown to increase overall survival for glioblastoma patients [18,19].

The significance of tumor BV during treatment has been assessed previously using DSC-MRI. Cao, Galban and co-workers at the University of Michigan found that changes in the mean values of BV for the whole tumor did not contain predictive information regarding treatment response [5,20]. This

group used an elegant but elaborate technique where voxel-by-voxel comparisons allowed for the generation of so-called parametric response maps (PRMs) that evaluated regional changes. It was shown that the percentage of voxels within the tumor with large changes in BV (both increases and decreases of more than 20%) correlated to outcomes. Other groups have successfully employed similar functional maps in the context of diffusion-weighted MRI [21]. In this study, we did not attempt to carry out voxel-to-voxel analysis for a number of reasons. Firstly, it requires a highly precise image registration that we believe may be difficult to achieve for all patients. Additionally, change in tumor morphology within the contoured contrast enhancing area further complicates a reliable voxel-to-voxel relationship, even when using a deformable registration approach. Furthermore, we believe that the subtraction of tumor/surgical cavities that we carried out in some respects minimizes the influence of 'dead' perfusion space on the median values used. Finally, the method we used generated voxels with a size of $3.1 \times 2.5 \times 8$ mm^3. Differences in responses dependent on the microenvironments may even exist within a voxel, which then itself only represents a weighted average of values.

Limitations of the study

We chose to measure parameters of vascular physiology within a static ROI defined at the baseline scan. This has the practical advantage of not requiring contouring of tumor for every examination. However, with this method there is a risk of the tumor moving out of the ROI due to either increased or decreased edema during RT, treatment response, or tumor growth with central cavitation. The tumor mask (ROI for measurement) was therefore visually inspected for each scan and found to be generally acceptable although in some cases, minor mismatches were visible. Another limitation of our method was the small vertical field-of-view (36.5 mm). As covering the whole tumor was prioritized in this study, in most cases (10/11) it was necessary to perform two bolus trackings ('runs'). For this reason, we chose to normalize the data to NAWM. It may be adequate to carry out only one run with the slice placed centrally in the tumor as this may be physiologically representative, but this needs to be validated.

As mentioned above, DCE-MRI created voxels measuring $3.1 \times 2.5 \times 8$ mm^3. This spatial resolution is far lower than that of standard structural (non-dynamic) MRI sequences and, thus, the method is susceptible to so-called partial volume effects (i.e. the signal from smaller volumes of interest is underestimated due to 'dilution' of signal from surrounding healthy tissue). This is a common limitation in metabolic imaging which also applies to PET [22] and single-photon emission computed tomography (SPECT) technologies. In this small study, we found no correlations between tumor size and vascular physiology parameters, but an underestimation in the case of the smallest tumor cannot be ruled out.

We unexpectedly found a significant decrease in the Ki of NAWM at MRI_4. It is unknown whether this was a random occurrence or an effect of having completed RT (which is known to induce a mild but temporary edema of the brain for some patients).

We have previously shown that the signal-to-noise ratio in a 1.5 T MRI scanner is too low to generate vascular physiology maps using DCE-MRI [6] which is a disadvantage of this technique.

Finally, the small number of patients as well as poor survival times makes it impossible to draw statistically sound conclusions regarding the prognostic value of measures of vascular physiology. Although we could not detect any differences in these parameters between patients with longer versus shorter PFS, this study cannot rule out that such a difference actually exists. A larger cohort of patients would be needed to draw such a conclusion. PFS-6 correlates reasonably well to overall survival [23], and was therefore chosen as the main clinical endpoint in this study. But four of the six patients with PFS longer than six months had experienced disease progression within 12 months of diagnosis and thus the biological differences between the groups we have compared may have been discrete, which also stresses the need for a larger study group.

In conclusion, using DCE-MRI at early intervals during radio-chemotherapy for glioblastoma, we have shown novel changes in tumor BF and we have described a distinct time-curve for parameters of vascular physiology. This included tumor Ki, which has not been assessed previously under these circumstances. We found no correlations to outcome but this should be investigated in a larger cohort of patients.

Declaration of interest: This work was funded by the Capital Region of Denmark and by the Danish Center for Interventional Research in Radiation Oncology. We are grateful to Adam Espe Hansen and Otto Henriksen for their expert advice during the entire study and to radiographers Helle Juhl Simonsen, Bente Sonne Møller and Marjut Lindhardt at the Functional Imaging Unit, Glostrup Hospital for assistance in carrying out all MRI. The authors report no conflicts of interest. The authors alone are responsible for the content and writing of the paper.

References

[1] Stupp R, Mason WP, van den Bent MJ, Weller M, Fisher B, Taphoorn MJ, et al. Radiotherapy plus concomitant and adjuvant temozolomide for glioblastoma. N Engl J Med 2005;352:987–96.

[2] Law M, Yang S, Wang H, Babb JS, Johnson G, Cha S, et al. Glioma grading: Sensitivity, specificity, and predictive values of perfusion MR imaging and proton MR spectroscopic imaging compared with conventional MR imaging. Am J Neuroradiol 2003;24:1989–98.

[3] Paulson ES, Schmainda KM. Comparison of dynamic susceptibility-weighted contrast-enhanced MR methods: Recommendations for measuring relative cerebral blood volume in brain tumors. Radiology 2008;249:601–13.

[4] Cao Y, Tsien CI, Nagesh V, Junck L, Ten Haken R, Ross BD, et al. Clinical investigation: Survival prediction in high-grade gliomas by MRI perfusion before and during early stage of RT. Int J Radiat Oncol Biol Phys 2006;64:876–85.

[5] Galbán C, Chenevert T, Meyer C. Response map-derived MRI biomarkers: Identification of early and distinct glioma response patterns not predicted by standard radiographic assessment. Clin Cancer Res 2011;17:4751–60.

[6] Larsson HB, Hansen AE, Berg HK, Rostrup E, Haraldseth O. Dynamic contrast-enhanced quantitative perfusion measurement of the brain using T1-weighted MRI at 3T. J Magn Reson Imaging 2008;27:754–62.

[7] Larsson HBW, Courivaud F, Rostrup E, Hansen AE. Measurement of brain perfusion, blood volume, and blood-brain barrier permeability, using dynamic contrast-enhanced T-1-weighted MRI at 3 Tesla. Magn Reson Med 2009; 62:1270–81.

[8] Jain R. Measurements of tumor vascular leakiness using DCE in brain tumors: Clinical applications. NMR Biomed 2013;26:1042–9.

[9] Cao Y, Tsien CI, Shen Z, Tatro DS, Ten Haken R, Kessler ML, et al. Use of magnetic resonance imaging to assess blood-brain/blood-glioma barrier opening during conformal radiotherapy. J Clin Oncol 2005;23:4127–36.

[10] Henriksen O, Larsson HBW, Hansen AE, Grüner JM, Law I, Rostrup E. Estimation of intersubject variability of cerebral blood flow measurements using MRI and positron emission tomography. J Magn Reson Imaging 2012;35:1290–9.

[11] Hansen AE, Pedersen H, Rostrup E, Larsson HBW. Partial volume effect (PVE) on the arterial input function (AIF) in T-1-weighted perfusion imaging and limitations of the multiplicative rescaling approach. Magn Reson Med 2009; 62:1055–9.

[12] Patlak CS, Blasberg RG, Fenstermacher JD. Graphical evaluation of blood-to-brain transfer constants from multiple-time uptake data. J Cereb Blood Flow Metab 1983;3:1–7.

[13] Jenkinson M, Bannister P, Brady M, Smith S. Improved optimization for the robust and accurate linear registration and motion correction of brain images. Neuroimage 2002;17:825–41.

[14] Brandsma D, Stalpers L, Taal W, Sminia P, van den Bent MJ. Clinical features, mechanisms, and management of pseudoprogression in malignant gliomas. Lancet Oncol 2008;9: 453–61.

[15] Gorski DH, Beckett MA, Jaskowiak NT, Calvin DP, Mauceri HJ, Salloum RM, et al. Blockade of the vascular endothelial growth factor stress response increases the antitumor effects of ionizing radiation advances in brief blockade of the vascular endothelial growth factor stress response increases the antitumor effects of ionizing radia. Cancer Res 1999;59:3374–8.

[16] Ellis LM, Hicklin DJ. VEGF-targeted therapy: Mechanisms of anti-tumour activity. Nat Rev Cancer 2008;8:579–91.

[17] Sorensen AG, Batchelor TT, Zhang W-T, Chen P-J, Yeo P, Wang M, et al. A "vascular normalization index" as potential mechanistic biomarker to predict survival after a single dose of cediranib in recurrent glioblastoma patients. Cancer Res 2009;69:5296–300.

[18] Gilbert MR, Dignam JJ, Armstrong TS, Wefel JS, Blumenthal DT, Vogelbaum MA, et al. A randomized trial of bevacizumab for newly diagnosed glioblastoma. N Engl J Med 2014;370:699–708.

[19] Chinot OL, Wick W, Mason W, Henriksson R, Saran F, Nishikawa R, et al. Bevacizumab plus radiotherapy-temozolomide for newly diagnosed glioblastoma. N Engl J Med 2014;370:709–22.

[20] Galbán C, Chenevert T, Meyer C. The parametric response map is an imaging biomarker for early cancer treatment outcome. Nat Med 2009;15:572–6.

[21] Ellingson B, Malkin M. Volumetric analysis of functional diffusion maps is a predictive imaging biomarker for cytotoxic and anti-angiogenic treatments in malignant gliomas. J Neurooncol 2011;102:95–103.

[22] Busk M, Horsman MR, Overgaard J. Resolution in PET hypoxia imaging: Voxel size matters. Acta Oncol 2008;47: 1201–10.

[23] Ballman K V, Buckner JC, Brown PD, Giannini C, Flynn PJ, LaPlant BR, et al. The relationship between six-month progression-free survival and 12-month overall survival end points for phase II trials in patients with glioblastoma multiforme. Neuro Oncol 2007;9:29–38.