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ACRIN 6684: Assessment of Tumor Hypoxia in Newly Diagnosed Glioblastoma Using $^{18}$F-FMISO PET and MRI

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Abstract

Purpose: Structural and functional alterations in tumor vasculature are thought to contribute to tumor hypoxia which is a primary driver of malignancy through its negative impact on the efficacy of radiation, immune surveillance, apoptosis, genomic stability, and accelerated angiogenesis. We performed a prospective, multicenter study to test the hypothesis that abnormal tumor vasculature and hypoxia, as measured with MRI and PET, will negatively impact survival in patients with newly diagnosed glioblastoma.

Experimental Design: Prior to the start of chemoradiation, patients with glioblastoma underwent MRI scans that included dynamic contrast enhanced and dynamic susceptibility contrast perfusion sequences to quantify tumor cerebral blood volume/flow (CBV/CFB) and vascular permeability ($K_{trans}$) as well as $^{18}$F-Fluoromisonidazole ($^{18}$F-FMISO) PET to quantify tumor hypoxia. ROC analysis and Cox regression models were used to determine the association of imaging variables with progression-free and overall survival.

Results: Fifty patients were enrolled of which 42 had evaluably imaging data. Higher pretreatment $^{18}$F-FMISO SUV$_{max}$ ($P = 0.048$), mean $K_{trans}$ ($P = 0.024$), and median $K_{trans}$ ($P = 0.045$) were significantly associated with shorter overall survival. Higher pretreatment median $K_{trans}$ ($P = 0.021$), normalized RCBV ($P = 0.0096$), and nCBF ($P = 0.038$) were significantly associated with shorter progression-free survival.

SUV$_{max}$ [AUC = 0.75; 95% confidence interval (CI), 0.59–0.91], RCBV [AUC = 0.72; 95% CI, 0.56–0.89], and nCBF [AUC = 0.72; 95% CI, 0.56–0.89] were predictive of survival at 1 year.

Conclusions: Increased tumor perfusion, vascular volume, vascular permeability, and hypoxia are negative prognostic markers in newly diagnosed patients with glioblastoma, and these important physiologic markers can be measured safely and reliably using MRI and $^{18}$F-FMISO PET. Clin Cancer Res; 22(20): 5079–86. ©2016 AACR.

Introduction

Despite significant research efforts and multimodal therapy with surgery, radiotherapy, and chemotherapy, glioblastoma remains the most aggressive and lethal primary malignant brain tumor (1). A major contributor to this treatment resistance is the abnormal tumor vasculature that is a pathologic hallmark of glioblastoma (2, 3). This abnormal vasculature is inefficient and heterogeneous resulting in overall poor delivery of oxygen leading to hypoxia and poor delivery of nutrients or cytotoxic chemotherapy to tumor tissue (4).

Hypoxic cancers are particularly challenging to treat for a variety of reasons. Hypoxia accelerates glycolysis and leads to an acidic microenvironment, which shifts macrophages to a protumor and immunosuppressive phenotype (5, 6). Normal cells undergo apoptosis when exposed to hypoxic conditions, and there is selection for more malignant, invasive cells and cells with a cancer “stem cell” phenotype (7, 8). Radiation needs oxygen to be most effective so hypoxic areas of tumor are resistant to radiotherapy (9). The identification of aberrantly functioning vasculature and hypoxic tumor tissue would be valuable in predicting tumor behavior and might help select treatments more likely to be effective in tumors with these features. Recent developments in functional and molecular imaging (specifically dynamic MRI and hypoxia PET imaging) can provide information about these physiologic changes and how they could influence patient management.

$^{18}$F-Fluoromisonidazole ($^{18}$F-FMISO) is a PET imaging agent that selectively binds to hypoxic tissues. $^{18}$F-FMISO is not
Translational Relevance

Glioblastomas are incurable, highly malignant tumors characterized by microvascular proliferation. These new tumor blood vessels are very inefficient, leading to areas of hypoxia and necrosis that contribute to treatment resistance because of decreased efficacy of radiation, poor delivery of chemotherapy, and negative effects on immunosurveillance. Considerable research has focused on targeting this abnormal tumor vasculature for therapeutic benefit, but trials to date have shown limited impact on improving overall survival likely because of lack of appropriate patient selection. Using MRI and 18F-Fluoromisonidazole PET (18F-FMISO PET), changes in tumor cerebral blood volume/flow, vascular permeability, and hypoxia can be measured noninvasively. Armed with this vital physiologic information, we will be able to design better trials of vascular and hypoxia targeting agents by selecting those patients most likely to benefit from this therapeutic avenue.

The treating physician) after surgery based on postcontrast MRI or fluid-attenuated inversion recovery (FLAIR) imaging, although the minimal amount was not specified. Other key eligibility criteria included Karnofsky performance status (KPS) >60 and age >18 years. Patients were followed every 3 months for tumor progression and survival for at least 1 year.

MRI and PET image acquisition

An MRI and an 18F-FMISO PET were performed after tumor diagnosis and within 2 weeks prior to initiation of chemoradiation. All sites had to follow a standardized acquisition protocol for these scans, and each site submitted a presubmission qualification MRI demonstrating their ability to complete the MRI as per protocol requirements. The MRI included DCE, DSC, and routine sequences (e.g., precontrast/postcontrast T1-weighted, diffusion-weighted imaging, and FLAIR images; see http://www.acrin.org/Portals/0/Protocols/6684/ACRIN6684_Amend7_012412_master_ForOnline.pdf for full details). For DCE data collection, the preinjection T1 data were obtained using a multiple flip angle T1-weighted SPGR technique (TR 4-6 ms, TE min, FA 2, 10, 15, 20, and 30 degrees). Next, dynamic T1-weighted SPGR images (TR 4-6 ms, TE min, FA 20 degrees) were obtained every 5.5 seconds, for a total imaging time 5.5 minutes during the first bolus injection of gadolinium contrast agent. This was followed by standard postcontrast imaging and then collection of the DSC data during which a second dose of gadolinium contrast was administered. This order of imaging was followed so that the contrast dose used for DCE imaging served as a preload for DSC imaging and serves to diminish confounding contrast agent leakage effects (17).

PET/CT scanners were also prequalified by ACRIN using a flood phantom and sample patient images, which required reconstruction with identical parameters as patient emission images. Scanners were required to undergo cross-calibration to a gamma well counter which was used for determination of blood sample activity concentration at each participating center. A 20-minute static 18F-FMISO PET emission image was acquired 110 minutes after injection of 3.7 MBq/kg of 18F-FMISO during which three venous blood draws at 5, 10, and 15 minutes postscan start were obtained for quantitative hypoxia analysis as described previously (13). Measures of radioactivity from the blood samples were cross-calibrated to the PET scanner at each center following a standardized procedure developed by ACRIN. A low-dose CT transmission scan provided attenuation mapping at the time of reconstruction. Image reconstruction included 3D filtered back projection and iterative techniques, where filter sizes ranged from 2 to 6 mm, and reconstructed image resolution varied from 1 to 6 mm.

All image analysis was performed by central laboratories with expertise in hypoxia PET, dynamic MRI, or diffusion MRI analysis. Specific analysis methods were described in the information to follow. The included patients were imaged at nine centers on Philips 3T (12 patients), GE 3T (12 patients), Siemens 3T (2 patients), and Siemens 1.5T (five patients) magnets.

MRI analysis

DCE MRI. DCE MRI analysis was performed by first computing the preinjection T1 map from the multiple flip angle T1-weighted SPGR data. This precontrast map was used to convert the signal intensities obtained from the dynamic acquisition into estimates of gadolinium contrast-concentrations over time (ΔR1(t)). Finally, parameter maps of ktrans were

Patients and Methods

Study design and patient selection

This study was conducted through the American College of Radiology Imaging Network (ACRIN) in patients with newly diagnosed glioblastoma (NCT009920577). The trial was approved by the institutional review boards at all participating sites. Prior to enrollment, all patients signed an informed consent document. Eligible patients had to have a histologic diagnosis of glioblastoma and a plan to receive standard radiation and temozolomide chemotherapy. In addition, patients could receive an investigational agent as part of a clinical trial.

Patients needed to have some residual tumor (as determined by

retrained in normoxic tissue but binds tissue at oxygen tension of approximately 2 to 3 mm Hg and below (10). 18F-FMISO trapping requires active nitroreductase enzymes so is not trapped in areas of necrosis or nonviable cells, which could be a confounder in glioblastoma imaging. 18F-FMISO has been extensively validated as a marker of tumor hypoxia in animal models and smaller human studies (11, 12). Single-institution studies found that 18F-FMISO uptake is common in glioblastoma and that a greater pretreatment tumor hypoxic volume (IV) was associated with decreased survival in patients with newly diagnosed with glioblastoma suggesting that 18F-FMISO PET may provide a tool for risk stratification in these patients (13, 14).

Tumor oxygenation is thought to be mediated by changes in cerebral blood flow (CBF), vascular density or blood volume, and microvessel geometry, all factors that can be measured using dynamic susceptibility contrast (DSC) and dynamic contrast-enhanced (DCE) MRI perfusion techniques (9, 15, 16). We conducted a multicenter, phase II study to determine whether 18F-FMISO PET and MRI could measure tumor hypoxia and changes in tumor vasculature to predict survival in patients with newly diagnosed glioblastoma. The primary aim was to determine the association of baseline hypoxia, as measured by 18F-FMISO PET, and vascular parameters, as measured by DSC/DCE MRI with overall survival (OS).
computed using a matrix-based linearization method to fit tissue ΔR1(t) to the extended Tofts model (18).

**DSC MRI.** The raw DSC MRI time series were truncated to eliminate the first 5 seconds of the time course allowing the MR signal to reach steady state. The truncated DSC MRI time courses, S(t), were converted into concentration-time (i.e., AR2*(t)) curves using the following equation:

$$ΔR2^*(t) = -\frac{1}{TE} \ln \frac{S(t)}{S_0}$$  \hspace{1cm} (1)

where $S_0$ is the mean baseline value (17). The RCBV maps, uncorrected for leakage effects, were computed from the integral of AR2*(t) described by equation (1). Next, the RCBV maps were corrected for leakage effects and normalized to normal-appearing white matter (nRCBV) using OsiriX-open-source software with the IB Neuroplug-in Imaging Biometrics, LLC; ref. 17. The CBF maps were also determined using IB Neuro. First, an arterial input function (AIF) was automatically determined as an average of three automatically determined individual AIFs. The CBF was then determined from the tissue residue function derived from the deconvolution of the AIF with AR2*(t) (19). For more consistent cross-patient comparisons, the CBF was normalized to the mean of the region of interest (ROI) in normal-appearing white matter to produce the nCBF.

**Diffusion MRI.** Apparent diffusion coefficient (ADC) maps were aligned and resampled to precontrast and postcontrast T1-weighted images. A double Gaussian mixed model was fit to the ADC histogram data using nonlinear regression in GraphPad Prism version 4.0c (GraphPad Software, Inc.). The double Gaussian mixed model was defined as $p(ADC) = N(m_{ADC}, \text{SD}_1) + (1 - f) \cdot N(m_{ADC}, \text{SD}_2)$, where $p(ADC)$ is the probability of obtaining a particular value of ADC in the histogram, $f$ is the relative proportion of voxels represented by the lower histogram, $N(m, \text{SD})$ represents a normal (Gaussian) distribution with mean $m$ and SD, and $s_1, \text{ADC}_1$ represents the lower, and $ADC_2$ represents the larger of the two Gaussian distributions. The accuracy of model fits was manually examined to exclude erroneous results. In some cases, nonlinear regression was rerun with different initial conditions until convergence was obtained between the model and ADC histogram data. The mean of the lower Gaussian curve, $m_{ADC}$, was used in subsequent analyses as previous studies have shown that $m_{ADC}$ is predictive of survival in various therapeutic scenarios (20-23).

**MRI regions of interest.** Contrast-enhancing ROI were determined using a semiautomatic AT1 method (provided by Imaging Biometrics, LLC) where-by a difference image was computed from standardized precontrast and standardized postcontrast T1-weighted images and a threshold applied to automatically determine the ROI (24). These ROIs were applied to the nRCBV, nCBF, $k^\text{max}$, and ADC maps from which the quantitative imaging parameters were extracted (25). When signal dropout occurs in the DSC data due to susceptibility effects near air/tissue or surgical cavity interfaces, for example, the ROI used for the DSC parameter analysis may be a subset of that used for the DCE and anatomic data.

**PET analysis.** $^{18}$F-FMISO images were decay corrected to the time of injection and converted to standardized uptake value (SUV) units by normalizing the emission image to the injected dose and patient weight. Conventional postcontrast T1-weighted images and FLAIR images acquired within 2 weeks of the PET scan were registered with the $^{18}$F-FMISO SUV images to aid in delineating the ROI. As FMISO uptake may occur outside the contrast-enhancing tumor region, ROIs encompassed the entire tumor volume by segmentation of the peritumoral abnormal FLAIR hyperintensity and were constructed using PMOD software (PMOD Tech.), after which they were applied to the FMISO SUV image. As FMISO is a freely diffusible tracer, the ROI was dilated to include regions of FMISO uptake beyond the FLAIR hyperintensity (26, 27). Tumor ROIs ranged from 10 to 237 cc, so we did not therefore perform partial volume correction. The FMISO image data were normalized by the average blood activity to produce pixel level tissue-to-blood ratio (T/B) values for all image slices. To quantify hypoxia in each tumor region, the pixel with the maximum T/B value ($T/B_{\text{max}}$) and the HV were determined, both of which are independent of the ROI size. The HV was determined as the average of pixels in the tumor ROI with a T/B ratio $\geq 1.2$ and was previously shown to indicate significant hypoxia (28). HV determines the spatial extent of hypoxia in a tumor, whereas $T/B_{\text{max}}$ reports the severity of hypoxia, both of which have been shown to be independent predictors of outcome in brain cancer (13, 27). Conventional quantitative PET parameters, $SU_{\text{max}}$ and $SU_{\text{peak}}$, were also extracted from the tumor regions, where $SU_{\text{peak}}$ was determined as the average SUV from a 1-cm circular ROI centered over the hottest pixel (cf. ACRIN 6668/RTOG 0235 protocol, page 21; http://www.acrin.org/Portals/0/Protocols/6668/ACRIN6668_Amend1_030210_ForONLINE.pdf).

**Statistical analysis.** The primary objective was to determine the association of baseline FMISO uptake parameters and vascular MRI parameters with OS in participants with newly diagnosed glioblastoma. Secondary aims included correlation of imaging parameters with time to progression, progression-free survival (PFS) at 9 months (PFS-9), and OS at 1 year (OS-1). The study was designed to enroll 46 evaluable participants to detect a log HR of 1.279 for $T/B_{\text{max}}$ with HV as a covariate with a 50% event rate to achieve 90% power when the type I error is set at 0.05. These parameters were selected on the basis of the best available preliminary data at the time of study design (13). Correlation between parameters was assessed using Pearson correlation. Kaplan–Meier survival estimates for time to death and PFS time were generated along with the median survival time with its 95% confidence interval (CI). The ROC curves for the imaging markers were constructed for two binary outcomes: OS-1 and PFS-9. The areas under the ROC curves (AUC) were estimated empirically with the trapezoid rule. The 95% CI of the AUCs were calculated using Delong method (29). A marker was considered effective in classification of an outcome status when its lower 95% CI of the AUC was at least 0.50. Each marker was modeled with a univariate Cox regression model for OS time and PFS time, separately. The HR, along with its 95% CI and the P value based on Wald statistic were reported. Analyses were done with SAS 9.4 and P < 0.05 was considered statistically significant. We have followed REMARK criteria in the reporting of this study (30).
Results

Patients' characteristics

Fifty patients with newly diagnosed glioblastoma were enrolled from 11 academic centers in the United States (Supplementary Table S1). All patients underwent initial diagnostic surgery (resection or biopsy), and the majority of patients were treated with standard involved field radiation with concomitant temozolomide followed by monthly maintenance temozolomide. Some patients also received additional therapy as part of a clinical trial.

Of the 50 patients enrolled, 42 (84%) had evaluable imaging studies, 38 patients had evaluable 18F-FMISO PET scans, 37 had evaluable DSC imaging, 31 had evaluable DCE imaging, and 29 had evaluable diffusion tensor imaging data. The most common reasons for incomplete imaging datasets included patient dropout prior to any imaging occurring and technical difficulty in 18F-FMISO production or MR sequence acquisition resulting in uninterpretable results because of incorrect acquisition parameters, insufficient contrast to noise, or poor contrast agent injection during DSC/DCE acquisition (Fig. 1). Patients tolerated the imaging well including the requirement for three venous blood draws during the 18F-FMISO PET scan, and there were no serious adverse events related to the imaging. Patient demographic information, tumor size, MGMT status, and treatment received for the 42 evaluable patients are presented in Table 1.

Imaging biomarkers

As our primary focus was tumor vasculature and hypoxia, we report on several variables that assessed these measures (Figs. 2 and 3). These variables included HV, 18F-FMISO SUV$_{peak}$, T$_{max}$, mean $k^{max}$, median $k^{max}$, nRCBV, and nCBF. As abnormal tumor vasculature is likely to influence hypoxia, we explored the correlation between vascular parameters and the P1T markers of tumor hypoxia. Most notably, there was a moderate positive correlation between nCBF and HV (0.5, P < 0.001). The correlations between other pairs of markers were not statistically significant.

PFS and OS

The median survival time for the 42 evaluable patients was 408 days (95% CI, 316–642), and 60% (25/42) of patients were alive at 1 year (OS-1). Similarly, the median PFS for the 42 evaluable

<table>
<thead>
<tr>
<th>Table 1. Patient cohort</th>
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<tr>
<td>Patient characteristics</td>
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<tr>
<td>Median age (range)</td>
</tr>
<tr>
<td>Gender, male (%)</td>
</tr>
<tr>
<td>Median residual CE tumor volume (range)$^b$</td>
</tr>
<tr>
<td>MGMT methylated</td>
</tr>
<tr>
<td>MGMT unmethylated</td>
</tr>
<tr>
<td>MGMT unknown/not tested$^b$</td>
</tr>
<tr>
<td>Median number of days from surgery to FMISO PET (SD)</td>
</tr>
<tr>
<td>Initial treatment</td>
</tr>
<tr>
<td>Temozolomide + RT</td>
</tr>
<tr>
<td>Temozolomide + RT + clinical trial drug</td>
</tr>
<tr>
<td>Other$^c$</td>
</tr>
<tr>
<td>Salvage therapy$^d$</td>
</tr>
<tr>
<td>None</td>
</tr>
<tr>
<td>RT</td>
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<tr>
<td>Surgery</td>
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<tr>
<td>Bevacizumab</td>
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<tr>
<td>Chemotherapy</td>
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<tr>
<td>NovoTTF</td>
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</tbody>
</table>

Survival

| Median survival time, days (95% CI) | 408 (316, 642) |
| Alive at 1 year (%)                 | 25 (60%)      |
| Median PFS, days (range)            | 258 (190, 335) |
| Progression free at 9 months (%)    | 79 (45%)      |

Abbreviations: CE, contrast enhancing; RT, radiotherapy. $^a$Central review of tissue was not required, and not all sites tested MGMT or patient underwent a biopsy so there was insufficient tissue for MGMT testing. $^b$Included bevacizumab alone, temozolomide alone, or temozolomide with BSI2CL. $^c$Patients could be counted more than once if received multiple salvage therapies.

patients was 258 days (95% CI, 190–335), and 45% (19/42) of the patients were progression-free at 9 months (PFS-9; Table 1).

ROC analysis

ROC analysis found SUV$_{peak}$ (AUC = 0.75; 95% CI, 0.59–0.91), nRCBV (AUC = 0.72; 95% CI, 0.56–0.89), and nCBF (AUC = 0.72; 95% CI, 0.56–0.89) were predictive of survival at 1 year with higher values predicting decreased likelihood of
survival at 1 year (Table 2). SUV_{max} has similar results to SUV_{peak}. None of the other markers was predictive of OS-1 or PFS-9 (Table 2).

**Association of imaging variables and survival measures**

In the univariate Cox model, higher ^18F-FMISO SUV_{peak} ($P = 0.048$), mean $k^{trans}$ ($P = 0.024$), and median $k^{trans}$ ($P = 0.043$) were significantly associated with shorter survival time. Only mean $k^{trans}$ maintained its significant association in a multivariate Cox model that included age and tumor volume. Higher median $k^{trans}$ ($P = 0.021$), nRCBV ($P = 0.0986$), and nCBF ($P = 0.038$) were significantly associated with shorter PFS, whereas higher mean $k^{trans}$ had borderline significance ($P = 0.074$; Table 3). Only nRCBV maintained its significant association with PFS in a multivariate Cox model that included age and tumor volume. None of the other markers had significant associations with PFS or OS.

**Serial imaging**

A subset of patients ($N = 4$) underwent serial imaging with FMISO PET at baseline, mid-radiation and 4 weeks after radiation. Although this was a small subset, there appeared to be a mixed response in HV during treatment with some tumors demonstrating increased HV and others demonstrating decreased HV during treatment (Supplementary Table S2). TB_{max} and SUV_{peak} demonstrated less variability over time. HV provides information about the spatial extent of hypoxia so is a marker of particular interest when assessing change in tumor hypoxia with treatment.

**Diffusion imaging**

To look at the impact tumor cell density might have on hypoxia, or as a standalone prognostic marker of survival, we evaluated diffusion characteristics ($m_{ADC}$) within the enhancing tumor. Baseline diffusion characteristics were not significantly associated with OS, suggesting diffusion characteristics of the tumor may not be prognostic biomarkers of survival.

**Discussion**

In this multicenter study, we found that pretreatment abnormal tumor vasculature (as measured by MRI) and hypoxia (as measured by ^18F-FMISO PET) were associated with worse survival in patients with newly diagnosed glioblastoma. These results build upon and extend prior studies highlighting the negative impact tumor hypoxia has on patient outcome (13, 31). Specifically, those tumors with the most elevated baseline markers of vascular permeability and perfusion ($k^{trans}$, nRCBV, nCBF) and hypoxia (peak ^18F-FMISO SUV) were associated with worse survival and there was a positive correlation between elevated nCBF and hypoxia (HV) suggesting that the abnormal tumor vasculature was contributing to tumor hypoxia. Thus, inefficient tumor perfusion and increased vascular permeability appear to be driving tumor malignancy and contributing to tumor hypoxia in newly diagnosed glioblastoma.

The increased nCBF at baseline in conjunction with poor oxygenation may indicate the tumor’s effort to promote angiogenesis in the face of hypoxia. Elevated $k^{trans}$ and nRCBV/nCBF also indicate an abnormal and inefficient tumor vasculature so may reflect the presence of arteriovenous shunts in high-grade glial tumors (32). This could explain why poorer prognosis is generally associated with higher baseline perfusion values and would support recent findings that heterogeneous flow and microvascular shunting may be more predictive of poor prognosis than CBV alone (33, 34). Although only a subset of the total...
patients, the change in HV in four patients who underwent serial FMISO PET varied possibly suggested different changes in vascular architecture with treatment but this needs to be studied in more detail in a larger patient population. Regional tumor hypoxia resulting from abnormal tumor vasculature has a negative impact on the efficacy of radiation, chemotherapy, immune surveillance, normal apoptosis, genomic stability, and angiogenesis (5). As our results demonstrate, we can now quantitate hypoxia with $^{18}$F-FMISO PET and alterations in tumor vasculature with DSC/DCE MRI to fully investigate how structural changes in blood vessels cause functional changes in the tumor microenvironment and specifically, tumor hypoxia.

Of the potential parameters reflecting tumor hypoxia in our study peak, $^{18}$F-FMISO SUV strongly predicted survival based on ROC analysis. Previous single-institution $^{18}$F-FMISO PET studies in newly diagnosed glioblastoma had found that HIV, the volume of hypoxic tumor below a particular threshold, was predictive of outcome (13, 14). Specific threshold values are harder to interpret when considering different centers and PET scanner types, and the smaller tumor volumes in this study compared with the prior published studies are more sensitive to partial volume effects and likely explains why SUV performed more robustly than HIV. Given that SUV is already a widely used tool for clinical $^{18}$F-FDG PET imaging, using SUV will be a more easily implementable approach. However, in light of previously published data the value of other $^{18}$F-FMISO PET measures such as HIV or SUV requires further validation.

In this patient group, imaging was obtained after surgery but before treatment with chemoradiation. Considerable literature supports the concept that extent of resection and/or residual tumor volume significantly influences survival (35, 36). In general, judgments about extent of resection and residual tumor are made on the basis of residual contrast-enhancing abnormality on imaging. However, this assessment is often limited because postsurgical contrast-enhancing abnormalities do not necessarily reflect neoplastic tissue and residual neoplasm does not always cause enhancement. The significant relationship between $k^{\text{trans}}$ in the residual tumor and survival suggests that $k^{\text{trans}}$ may be an independent indicator of residual disease and poor survival in this patient population. One possible explanation of this finding would be that residual enhancement due to postsurgical granulation tissue is associated with a lower $k^{\text{trans}}$ than enhancement due to residual neoplasm. Thus, $k^{\text{trans}}$ may be more useful than residual tumor area in prognostication.

There were several limitations to our study. Not all 50 anticipated patients ended up participating in all the imaging studies, reducing the total available for final analysis. Further studies in larger populations will need to be done to confirm our findings.
Table 3. Cox regression model

<table>
<thead>
<tr>
<th>Parameter</th>
<th>Overall survival time</th>
<th>Progression-free survival</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>HR 95% CI</td>
<td>HR 95% CI</td>
</tr>
<tr>
<td>Univariate model</td>
<td></td>
<td></td>
</tr>
<tr>
<td>SUVmax</td>
<td>1.54 1.00–2.36 0.048p</td>
<td>1.24 0.80–1.91 0.33</td>
</tr>
<tr>
<td>TBmax</td>
<td>1.16 0.75–1.81 0.50</td>
<td>0.93 0.61–1.40 0.72</td>
</tr>
<tr>
<td>HV</td>
<td>1.00 0.97–1.03 0.90</td>
<td>1.01 0.98–1.04 0.36</td>
</tr>
<tr>
<td>Mean Ktrans, a</td>
<td>1.17 1.00–1.34 0.024p</td>
<td>1.10 0.99–1.23 0.074</td>
</tr>
<tr>
<td>Median Ktrans, a</td>
<td>1.32 1.01–1.72 0.045p</td>
<td>1.30 1.04–1.63 0.021p</td>
</tr>
<tr>
<td>nRCBV</td>
<td>1.11 0.90–1.37 0.31</td>
<td>1.18 1.06–1.54 0.0096p</td>
</tr>
<tr>
<td>nCBF</td>
<td>1.07 0.88–1.29 0.51</td>
<td>1.18 1.01–1.38 0.038p</td>
</tr>
</tbody>
</table>

*p<0.01 increase.

Although a key outcome of this study is the demonstration of the ability to conduct a multicenter trial with both advanced MRI and an investigational PET agent, a key lesson from the trial is that imaging complexity increases the rate of invaluable imaging data. Patients underwent variable extents of resection, and as we did not specify a minimum amount of residual tumor required. This resulted in some patients having very small residual enhancing tumors, which limited the analysis of the DSC and DCE MRI results, as these were based on enhancing tumor volume but should not have affected the interpretation of the FMISO scans, as FMISO is not limited by the BBB. Finally, the treatment regimen was not standardized, and MGMT methylation status, a known prognostic marker, could not be determined in all patients.

In summary, increased pretreatment tumor nRCBV, nCBF, and vascular permeability (Ktrans) as well as increased tumor hypoxia were associated with poorer survival in patients with newly diagnosed glioblastoma. Using a combination of 18F-FMISO PET and MRI, these markers of abnormal tumor vasculature can be measured noninvasively to provide prognostic information and help identify those patients most at risk for early tumor progression. The lack of a strong correlation between most of the PET and MRI parameters is suggestive of the unique and complementary role that each may play for assessing tumor status. Overall, knowledge about tumor vascular structure and function may be useful in radiation planning where hypoxia impacts radiation efficacy, optimization of antiangiogenic therapy which can modulate vasculature and resultant areas of hypoxia, and guiding the development of hypoxia targeting drugs for patients with glioblastoma as well as other cancer types (37). This prospective, multicenter study supports the utility of these imaging approaches in newly diagnosed glioblastoma and warrants further study in a larger population, particularly if coupled to hypoxia-directed therapy.

Disclosure of Potential Conflicts of Interest

K. Schmaida has ownership interest (including patents) in Imaging Biometrics LLC. D. Barboriak is a consultant/advisory board member for GE Medical Systems. D.A. Mankoff is a consultant/advisory board member for GE Healthcare and reports receiving commercial research grants from Siemens Healthcare. No potential conflicts of interest were disclosed by the other authors.

Authors’ Contributions

Conception and design. E. Gerstner, Z. Zhang, L. Hanna, K. Schmaida, A.G. Sorensen, D.A. Mankoff


Acquisition of data (provided animals, acquired and managed patients, provided facilities, etc.). E. Gerstner, J. Fink, M. Muiz, L. Hanna, A. Mintz, L. Rostakoglou, E.A. Eikman, A.G. Sorensen, D.A. Mankoff

Analysis and interpretation of data (e.g., statistical analysis, biostatistics, computational analysis). E. Gerstner, Z. Zhang, J. Fink, M. Muiz, L. Hanna, E. Greco, M. Prah, K. Schmaida, B.M. Ellington, E.-M. Rataj, D. Barboriak, D.A. Mankoff

Administrative, technical, or material support (i.e., reporting or organizing data, constructing databases). E. Gerstner, L. Hanna, M. Prah, D. Barboriak

Study supervision. E. Gerstner, J. Fink, A.G. Sorensen, D.A. Mankoff

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References

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