Diagnostic utility of Restriction Spectrum Imaging in the characterization of the peritumoral brain zone in glioblastoma: Analysis of overall and progression-free survival

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ABSTRACT

Purpose: We studied the ability of Restriction Spectrum Imaging (RSI), a novel advanced diffusion imaging technique, to estimate levels of cellularity in different glioblastoma regions, evaluated their prognostic value compared with established clinical diffusion metrics such as fractional anisotropy (FA) and mean diffusivity (MD).

Methods: Forty-two patients with untreated glioblastoma, IDH-wildtype, were examined with an advanced MRI tumor protocol. The region of interest (ROI) was obtained from the contrast-enhancing part of tumor and the peritumoral brain zones and then co-registered with RSI-cellularity index, FA and MD maps. Histogram parameters of diffusion metrics were assessed for all ROI locations and compared to MGMT promoter methylation status and survival. The ability of RSI-cellularity index, FA, and MD to stratify survival and were assessed by Cox proportional hazard regression, adjusted for significant clinical predictors.

Results: The highest RSI-cellularity index was measured in contrast-enhancing tumor core with a negative gradient from tumor core to the periphery of peritumoral zone with predictive accuracy 81 % (P < 0.001). Shorter overall survival was significant associated with higher RSI-cellularity index (hazard ratio (HR) 3.6, 95 % confidence interval (CI) 1.3–9.5, P = 0.002) with synchronal decrease in MD (HR 0.31, 95 %CI 0.1–0.8, P = 0.008) in the contrast-enhanced tumor core. This association was also consistent for RSI-cellularity index value measured in the peri-enhancing zone (HR 3.6, 95 % CI 1.0–12.3, P = 0.041). No statistically significant differences were noted between RSI-cellularity index, FA, nor MD and MGMT promoter methylation.

Conclusion: RSI-cellularity index may be used as prognostic biomarker to improve risk stratification in patients with glioblastoma.

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Overall survival
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1. Introduction

Glioblastoma is the most frequent malignant primary brain tumor in the adult population. Despite recent advances in treatment options, the prognosis of glioblastoma remains nearly uniformly fatal. Among other prognostic predictors, surgical treatment related to its extension is the only one that can be directly influenced in the initial stage of disease [1].

Complete macroscopic tumor removal (gross total resection) in patients with glioblastoma is a primary surgical goal and in comparison to sub-total resection prolongs overall survival (OS) and progression free survival (PFS) [2].

Glioblastoma growth is characterized by diffuse infiltration of normal brain tissue, with tumor cells moving through the hyaluronic acid-rich parenchyma towards microvasculature and then migrating...
rapidly along vascular tracks [3]. Intraoperative navigation, based mainly on postcontrast MRI, is the standard of care for glioma surgery in an effort to achieve maximal safety resection. Still, this technique is based on visual identification of regions with disrupted blood-brain barrier that cannot accurately detect tumor infiltration beyond the apparent borders of the enhancing region. Similarly, the current radiation treatment regimen involves exposure of a margin around the resection cavity, which normally receives a spatially uniform radiation dose [4,5]. Non-invasive and robust diagnostic biomarkers are therefore warranted for identification of high-cellularity components in order to increase cytoreductive treatment with real benefits to prognosis.

ADC derived from DWI is inversely related to tissue cellularity and has been proposed to be a noninvasive imaging biomarker for detection of intratumoral high cell density areas [6]. However, edema associated with inflammation, infiltration and necrosis increases extracellular diffusion and thus ADC values [7].

In this work we attempted to evaluate the role of Restriction Spectrum Imaging (RSI), a novel diffusion-weighted MRI technique, that separates the relative contributions of hindered and restricted signals originating from extracellular and intracellular water compartments, respectively, by using a multi-shell acquisition, together with an advanced linear mixture model to resolve a spectrum of length scales and incorporating geometric information [8,9]. Use of RSI has previously demonstrated to improve tumor delineation and also to help stratify survival in patients with glioblastoma [8,10,11].

The aims of this study were threefold: (1) to evaluate the ability of RSI for improved delineation of tumor core from peritumoral brain zone and perilesional normal-appearing zone; (2) to evaluate the association between diffusion metrics and O6-methylguanine-DNA methyltransferase (MGMT) status; (3) to investigate the association of RSI parameter with OS and PFS in patients with glioblastoma and whether the RSI-cellularity index value would be a more robust imaging biomarker than diffusion tensor imaging (DTI) derived parameters.

2. Materials and methods

2.1. Study design and ethics

This study is a prospective analysis of data acquired from Oslo University Hospital from June 2016 to December 2018. All patients provided written informed consent for use of clinical data and imaging surveys for research purpose. Institutional and regional medical ethics committees approved this study, REC-number 2018/2464.

2.2. Patient population

The prospective cohort included ninety-two patients who required an image-guided stereotactic biopsy or surgery for presumed high-grade gliomas referred from local hospitals. Fig. 1 shows the patient selection and dichotomization in a flow diagram with inclusion and exclusion criteria. Advanced tumor MRI examination (anatomic, diffusion MRI, perfusion MRI, spectroscopy and RSI imaging modalities) was obtained from all patients in our institution before surgical treatment. The diagnosis was based on histological and molecular examination of specimens obtained by stereotactic navigated biopsy or from tumor resection. In the final study cohort, forty-two patients with glioblastoma, IDH-wild-type, classified according to the 2016 World Health Organization (WHO) classification of tumors of the Central Nervous System (CNS) were included [12]. Maximum safe resection was performed by using neuronavigation (BrainLab) and 5-aminolevulinic acid (5-ALA) fluorescence guidance. The extension of resection was considered either gross total resection or subtotal resection [13]. Standard treatment, consisting of radiotherapy (a total 60 Gy in fractions of 2 Gy per day over 6 weeks) and concomitant/adjuvant chemotherapy (Temozolomide) was initiated 3–4 weeks after surgery. Patients and tumors characteristics

![Flow diagram demonstrates the patient selection and dichotomization with inclusion and exclusion criteria.](image-url)
including age, Karnofsky performance status, MGMT promoter methylation status and degree of surgical extent of resection are shown in Table 1.

### 2.3. Data acquisition/MRI protocol

All examinations were performed on a 3 T MR scanners (Skyra, Siemens Healthcare, Erlangen Germany) using a 20 channel head/neck coil. The following tumor protocol was carried out before gadolinium-based contrast agent (Clariscan 279.3 mg/mL, 0.2 mL/kg bodyweight, GE Healthcare, USA) administration: RSI, DTI, three-dimensional T1-weighted magnetization-prepared rapid gradient-echo (MPRAGE), fluid attenuated inversion recovery (FLAIR) and axial T2-weighted sequences. After contrast agent injection: dynamic susceptibility contrast perfusion MRI (DSC MRI) with a bolus injection (3 ml/s) of the Gadolinium-based contrast agent, followed by 30 ml of physiologic saline solution; susceptibility weighted MRI (SWI); three-dimensional T1-perfusion MRI (DSC MRI) with a bolus injection (3 ml/s) of the contrast agent. The descriptive diffusion parameters fractional anisotropy (FA) and mean diffusivity (MD) were derived based on the anisotropic DW data using weighted linear least-squares fit [18] and the b-values of 0, 200, 800, 1500 and 3000 s/mm² and with 12 directions at each respective nonzero b-value [8].

### 2.4. DNA extraction and molecular analyses

The DNA was extracted using the Maxwell 16 extractor (Promega, Madison, WI, USA) and the Maxwell 16 Tissue DNA Purification kit (Promega) according to the manufacturer’s recommendations. The mutational analyses were performed using M13-linked Polymerase chain reaction (PCR) primers designed to flank and amplify targeted sequences. All PCR were run on a Bio-Rad C100 Thermal Cycler (Bio-Rad Laboratories, Hercules, CA, USA). Direct sequencing was performed using a 3500 Genetic Analyser (Applied Biosystems, Foster City, CA, USA). The BLAST (http://blast.ncbi.nlm.nih.gov/blast.cgi) and BLAT (http://genome.ucsc.edu/cgi-bin/hgblat) programs were used for computer analysis of sequence data. Detailed information about analysis of IDH1, IDH2, TERT and MGMT using PCR is outlined in Supplementary Table 2 and were described previously [14].

### 2.5. Image processing and model estimation

All image postprocessing was carried out using the FMRIB Software Library v6.0 (FSL) [15] and Matlab R2018b (MathWorks, Natick, Massachusetts). First, the DWI were denoised using local Principal Component Analysis (LPCA) filter [16] and Gibbs ringing artifacts were removed [17]. Brain extraction, EPI-distortion, eddy-current and movement corrections were then applied using the bet, topup and eddy toolboxes from FSL. The quality of all corrected output data was evaluated by visual inspection. A standard tensor model was fit to the DW-data using weighted linear least-squares fit [18] and the b-values of 0, 200, 800 s/mm². The descriptive diffusion parameters fractional anisotropy (FA) and mean diffusivity (MD) were derived based on the tensor information [18]. Furthermore, RSI was estimated from the high-angular DW data based on the parameterization of the fiber orientation density function (FOD) using 4th order spherical harmonics, combined with an axially-symmetric Gaussian model [8]. The RSI spectrum was defined as a combination of anisotropic, restricted and free/hindered diffusion compartments as detailed by White et al. [8]. For the purpose of this study, we focused the analysis on the water signal fraction from the spherically restricted diffusion compartment. Based on the water signal fraction of this compartment, we generated RSI-cellularity index maps, where white and grey matter regions of apparent healthy brain tissue of 15 subjects were used as reference [9].

### 2.6. Segmentation of regions of interest

Tumor segmentation was performed semi-automatically based on region growing algorithm, on 3D volumetric T1 postcontrast sequence using a dedicated software package nordiCCE Version 4.0 (NordicNeuroLab AS, Bergen, Norway) by A.L. in collaboration with A.S., board-certified neuroradiologists (with 5 and 25 years of neuro-oncology imaging experience). Discrepancies were resolved through a consensus discussion. Both neuroradiologists were blinded for survival data. The volumetric regions of interest (ROI) were obtained from the contrast-enhancing tumor core (CET) in all cases. The peritumoral brain zone (PBZ) was defined as the area surrounding the CET in the absence of contrast-enhancement in the three-dimensional T1 magnetization-prepared rapid gradient-echo (MPRAGE). In addition, this area showed hyperintense signal in T2-weighted and FLAIR images [19]. ROI with extended margins were generated by means of morphological dilation of the CET using a spherical structuring element Matlab (v. R2017a, MathWorks Inc., Mass., USA). The peri-enhancing zone (PEZ) defined as the area of non-contrast enhancing tumor surrounding the CET at a distance of 5 mm. The near zone (NZ) defined between 5 and 10 mm from the CET and the far zone (FZ) defined between 10 and 15 mm from the CET. ROIs were also drawn in the ipsilateral normal-appearing zone (iNAZ) defined as the area immediately adjacent to the distal edge of the PBZ in and contralateral white matter to the tumor (cWMZ). The areas of necrosis were not included in further analysis. All ROI’s were then co-registered with the RSI-cellularity index, FA and MD respectively maps. Whole-tumor normalized histogram distributions of the RSI-cellularity index, FA and MD were created as described elsewhere [20]. Based on previous studies, MD 10th percentile and RSI-cellularity index 90th percentile values were used for further analysis [11]. The image postprocessing workflow is demonstrated in Fig. 2.

### 2.7. Survival assessment

Follow-up scans were acquired within 24 h after surgery, after radiotherapy and at 3-month intervals thereafter. OS and PFS were defined as time interval (number of days) from initial diagnosis to patient death or tumor progression, respectively. Tumor progression was defined according to updated Response Assessment in Neuro-Oncology criteria [21]. Patients still alive or lost to follow-up were censored for PFS on the date of their last central nervous system imaging study, and for OS on the date of their last clinical follow-up visit.

### 2.8. Statistical analysis

Analysis was performed using SPSS 25 (SPSS, Chicago, USA) and Stata 14 (STATA Corp., Texas, USA). The Mann-Whitney U test was used to compare diffusion parameters between different ROI inside and outside contrast enhanced tumor core. The diagnostic accuracy of this classification was also estimated by a classification and leave-one-out cross-validation (LOOCV) algorithm with 10-fold. The independent significance of various predictors (age, extent of resection, Karnofsky performance status scale and genetic profile) on OS and PFS were assessed by Univariate Cox proportional hazard regression analysis (CPH). Multivariate CPH analysis, that included significant clinical

### Table 1: Molecular profile and clinical characteristics of the patients sample.

<table>
<thead>
<tr>
<th>Gender²</th>
<th>Karnofsky performance status scale³</th>
<th>Surgical extension⁴</th>
<th>MGMT promoter methylation status⁵</th>
</tr>
</thead>
<tbody>
<tr>
<td>26/16 (62.8/37.2 %)</td>
<td>20/22 (47.6/52.4 %)</td>
<td>18/24 (42.9/57.1 %)</td>
<td>26/16 (61.9/38.1 %)</td>
</tr>
</tbody>
</table>

² No. of male/female (%).
³ No. of patients with Karnofsky performance scale ≥ 70/ No. of patients with Karnofsky performance scale < 70.
⁴ No. of subtotal resections/ No. of gross total resections.
⁵ No. of patients with MGMT methylated tumors/No. of patients with non-MGMT methylated tumors.
variables, was performed to evaluate the contribution of diffusion parameters from different tumor compartments to PFS and OS. ROC curves analysis was used to calculate optimal cut-off for dichotomize RSI-cellularity index, FA and MD metrics. For all cases, a two-tailed $P$-value of 0.05 or less was considered statistically significant.

3. Results

3.1. Patient characteristics and survival

The median age of the patients with glioblastoma was 65 years (range, 39–87 years). The Median PFS was 184 days, and the median OS was 245 days. Twenty-nine (69 %) patients had tumor progression following Response Assessment in Neuro-Oncology criteria and nine (21 %) were deceased at the end of the study inclusion time. CPH models revealed that the total resection and Karnofsky performance status over 70 were significantly associated with longer PFS (hazard ratio (HR) 3.5, 95 % confidence interval (CI) 1.0–12.1; $P = 0.04$ and HR 4.4, 95 %CI 1.6–12.1, $P = 0.006$) respectively, and longer OS (HR 3.8, 95 %CI 1.1–12.9, $P = 0.03$ and HR 3.2, 95 %CI 1.3–7.8, $P = 0.01$) respectively. These variables were included in subsequent multivariate CPH analyses.

3.2. RSI-cellularity index, FA and MD in different glioblastoma compartments

The RSI-cellularity index in CET was on average 24 % higher than the value in PEZ (median) ($P < 0.001$) and 28 % higher than in NZ ($P = 0.003$) (CET > PEZ > NZ). The corresponding predictive accuracy, obtained from LOOCV analysis were 81 % and 75 %, respectively. RSI-cellularity index in FZ retained a higher value, but not significant, compared to those of CET, PEZ and NZ. The lowest MD was estimated in CET with increasing values from CET to inNAZ (CET > PEZ > NZ), but significant differences between any parts of the PBZ were not observed. Moreover, the 25 % lower FA was measured in CET compared with PEZ with a diagnostic accuracy of 82 % ($P < 0.01$) (CET > PEZ). However, notable differences were not registered between FA values in CET and NZ or FZ. Scatter plots in Fig. 3 show the distribution of diffusion parameters values for all tumoral and peritumoral zones.

3.3. Relationship between survival outcome and diffusion metrics

Multivariate CPH models revealed that higher values of RSI-cellularity index with synchronal lower MD in CET were significantly associated with shorter PFS (177 vs 410 days, $p = 0.015$; 174 vs 378 days, $p = 0.029$, respectively) and OS (543 vs 215 days, $p = 0.002$ 173 vs 507 days, $p = 0.008$ respectively). This association was also consistent for RSI-cellularity index value measured in PEZ (PFS: 188 vs 429, $p < 0.048$ and OS: 298 vs 572, $p = 0.041$) and NZ (PFS: 150 vs 459, $p < 0.015$ and OS: 166 vs 643, $p < 0.020$). Lower FA values in CET and PEZ were significantly associated with poorer PFS (216 vs 441 days, $p = 0.006$) and OS (316 vs 601 days, $p = 0.006$). In contrast, no statistical significance was reached between MD values from PEZ, NZ, FZ and

Fig. 2. Image post processing workflow. The volumetric region of interest (ROI) was obtained from the contrast-enhancing tumor core and three peritumoral zones. All ROI’s were then co-registered with the RSI-cellularity index, FA and MD respective maps and normalized histogram distributions were created. Note: *MD-map, ** RSI-cellularity index map.

Fig. 3. Scatter plot with distribution (median with interquartile range) of a) RSI-cellularity index b) FA and c) MD values in CET, PEZ, NZ, FZ, inNAZ and cNAZ encoded by the colors in patients with glioblastoma. The RSI-cellularity index in CET was significantly higher than in PEZ ($P < 0.001$) and NZ ($P = 0.003$). Significantly lower FA was measured in CET ($P < 0.01$) without notable difference between FA values outside CET. No statistical difference was found between MD values in any of zones. The asterisks show significant differences.
survival outcome.

The total results of multivariate CPH analyses adjusting for significant clinical covariates (extension of resection, Karnofsky performance status scale) for diffusion image metrics are summarized in Table 2. The most significant survival predictors were RSI-cellularity index in CET, PEZ, NZ and FA in CET with respective hazard ratios of 3.60, 3.60, 3.0, and 5.72. The corresponding Kaplan-Meier survival curves for RSI-cellularity index with cut-off points and log-rank values for each of glioblastomas zones are shown in Fig. 4. Similar survival curves for FA and MD are shown in Figs. 5 and 6.

3.4. MGMT promoter methylation status and diffusion metrics

Presence of MGMT promoter methylation was significant associated with longer PFS (HR 3.3, 95% CI 1.4–7.9, P = 0.006) and longer OS (HR 3.7, 95% CI 1.6–8.7, P = 0.002). No statistically significant differences were noted between RSI-cellularity index, FA, nor MD and MGMT promoter methylation status obtained from CET and PBZ.

4. Discussion

The highest RSI-cellularity index was measured in CET with a negative gradient from tumor core to the periphery of peritumoral zone (PEZ and NZ). In contrast, we did not find significant differences between MD values in any parts of the peritumoral zones. This finding is supported by a previous study that showed the ability of RSI to improve tumor infiltration conspicuity and delineation from normal-appearing white matter compared with ADC [22]. The highest cellularity in CET may be explained by the dominant invasion and consequently highest density of glioma cells in perivascular space, leading to a disruption of endothelial tight junctions and leakage of contrast agent in CET [23,24].

Table 2

<table>
<thead>
<tr>
<th>Region of interest</th>
<th>Progression – free survival</th>
<th>Overall survival</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>P-value</td>
<td>HR (95% CI)</td>
</tr>
<tr>
<td>FA</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Peri-enhancing zone</td>
<td>0.140</td>
<td>0.21</td>
</tr>
<tr>
<td>Near zone</td>
<td>0.416</td>
<td>1.41 (0.5–3.3)</td>
</tr>
<tr>
<td>Far zone</td>
<td>0.193</td>
<td>0.63 (0.3–1.5)</td>
</tr>
<tr>
<td>RSI-cellularity index</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Peri-enhancing zone</td>
<td>0.015</td>
<td>3.48 (1.2–9.5)</td>
</tr>
<tr>
<td>Near zone</td>
<td>0.048</td>
<td>3.60 (1.0–12.3)</td>
</tr>
<tr>
<td>Far zone</td>
<td>0.015</td>
<td>4.70 (1.3–16.5)</td>
</tr>
<tr>
<td>Contrast-enhanced core</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Peri-enhancing zone</td>
<td>0.112</td>
<td>5.43 (1.2–16.9)</td>
</tr>
<tr>
<td>Contrast-enhanced tumor core</td>
<td>0.36</td>
<td>3.60</td>
</tr>
<tr>
<td>Near zone</td>
<td>0.029</td>
<td>0.37 (0.1–0.9)</td>
</tr>
<tr>
<td>Far zone</td>
<td>0.301</td>
<td>0.63 (0.3–1.5)</td>
</tr>
<tr>
<td>MD</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Peri-enhancing zone</td>
<td>0.722</td>
<td>0.85 (0.3–2.1)</td>
</tr>
<tr>
<td>Near zone</td>
<td>0.871</td>
<td>1.07 (0.4–2.6)</td>
</tr>
<tr>
<td>Far zone</td>
<td></td>
<td></td>
</tr>
</tbody>
</table>

HR = hazard ratio, RSI-cellularity index = restriction spectrum imaging -cellularity index, CI = confident interval, MD = mean diffusivity.

The decline of glioma cellularity from tumor core to periphery reflects an extensively infiltrating pattern in glioblastomas with the highest concentration of invading cells around primary tumor mass [25].

Cellularity in peritumoral zone has been investigated in several studies, in which the MRI findings were corroborated by histopathologic analyses, following stereotactic biopsy obtained from the peritumoral zone. The results of these studies are contradictory [26–29]. Barajas et al. investigated 119 tissue specimens and reported that ADC was associated with tumoral cellularity in the non-enhancing tumor component, but this was not the case in the contrast-enhanced component of the lesion [27]. Chang et al. [28] found that tumor cellularity was inversely correlated with ADC signal by using a voxel-level multiparametric MR imaging model, supporting the theory that water diffusion is restricted in hypercellular neoplastic environments. In contrast, Sadeghi et al. [29] did not find negative correlation between ADC ratios and cell density in either tumor core or the peritumoral tissue in examination of the 33 tumor tissue specimens. Discordance between findings may be explained by the inclusion in this study population of both low and high-grade gliomas [29], while Barajas [27] and Chang [28] investigated exclusively glioblastoma. The differences in strategies for obtaining tissue samples (targeted by contrast T1weighted [28,29] vs by diffusion and perfusion signal [27]) may be an additional reason for discordances.

FA, which reflects integrity and disruption of white matter fibers, was significantly lower in the CET than PEZ. This finding has been supported by previous studies demonstrating the most extensive loss of fiber connectivity in the aggressive parts of the tumor with highest cellularity [30,31]. Less variability was found between FA values in PEZ, NZ and FZ than it was observed between RSI-cellularity index values in the same zones. This finding may indicate that FA value is more severely affected than RSI-cellularity index by extracellular edema in peritumoral zone. Additional explanation may also be found in the unique invasion pattern that dominates in glioblastoma, tumor cells may migrate along white matter tracks and perivascular spaces [25]. Therefore, despite the differences of cellularity in peri-enhancing areas, the level of diffusion anisotropy is not necessary affected to the same degree.

Shorter survival outcomes were significantly associated with higher RSI-cellularity index in the CET. Interestingly, this correlation was also persistent in the PEZ and NZ. Thus, our findings highlight the importance of the peritumoral areas (PEZ and NZ) in order to predict further tumor expansion and tumor relapse [32]. We did observe a significant association between lower MD in the CET and shorter survival, but in contrast to RSI-cellularity index, MD in the peritumoral zones did not show predictive value to survival outcome. There is still no consensus in literature on how strong relation between the ADC value and tumor cellularity is. Indeed, several studies demonstrated the correlation between ADC and cellularity and, as consequence, also between ADC and survival outcome in patients with gliomas [33–35]. However, these correlations cannot be generalized. Based on the studies from Eidel et al. [36] and Stecco et al. [37], a weak-to-moderate inverse correlation was detected including only patients with glioblastoma. Possible explanation for these discrepancies may be found in the high heterogeneity of glioblastoma tissue. Theoretically, any modification of the extracellular matrix can influence the ADC. Glioma cells tend to produce large amount of extracellular matrix component [38], which serves as substrate for cells migration. These structures may exert considerable restriction for diffusion. On the other hand, destruction of normal extracellular matrix by glioma cells and secondary edema increases free diffusion and subsequent ADC. In addition, necrosis in different stages of microstructural tissue damage can lead to lower and elevated ADC values [39]. In agreement with previous studies, a reduction of FA in both CET and PEZ correlated with shorter survival time [31,40]. In contrast to RSI-cellularity index, FA values in both NZ and FZ had no apparent association to survival, which again highlights the potential of RSI to evaluate tumoral heterogeneity.
Thus, similar to a previous study, our results suggest that RSI may overcome some of the limitations of ADC and FA due to the reduction of sensitivity to the extracellular water [41]. RSI may therefore also provide a more reliable measure of increased cellularity associated with tumor progression.

Several studies have reported that MGMT promotor methylated tumors display diffusion MRI features of lower cellularity (high ADC) and severe disruption of white matter tracks (low FA) [42–44]. In contrast, we did not find significant association between any of diffusion metrics and methylation status. These conflicting results may be partially attributed to different methodology (3D ROI analyzed of whole volume tumor in our study versus 2D ROI of the solid part of tumor) [43,44] and different studies populations (IDH-wild type glioblastomas versus both IDH-mutant and IDH-wild type glioblastomas) [44,45]. In accordance with our results, Ahn S.S. et al. [46] and Gupta A. et al. [47] did not find any significant correlation between ADC/FA values and MGMT promotor methylation status.

Our work has some limitations. First, our data were collected from a single institution. Although this approach was selected in order to obtain a study in which all patients were examined on the same MR scanner with the same tumor protocol. In the future, a cross-site study including data from multiple medical centers needs to evaluate the reproducibility of our results. We have used PFS and OS as indicators to more infiltrative cell-rich regions with aggressive behavior. However, the lack of a histopathologic verification from the peritumoral brain zones is another important limitation. Furthermore, segmentation of the glioblastomas was performed semi-automatically that potentially can be a source for sampling bias. However region-grow algorithm for delineation of peritumoral region of interest and extraction of non-brain parenchymal structures was performed automatically. Finally, standard treatment, consisting of radiotherapy and concomitant chemotherapy was started within 4 weeks after surgery. However some differences in radiation doses and timing between radiotherapy sessions were observed, mainly related to patients with reduced health status.
5. Conclusion

The main findings of this prospective study was that higher RSI-cellularity index in the CET, but particularly also in the PEZ and NZ was associated with shorter survival outcomes, likely reflecting the potential of RSI to detect differences between the tumoral zones of glioblastomas. This characterization of glioblastoma compartments with different levels of cellularity, supplemented by other MRI sequences, can be helpful to optimize the extent of surgical resection, guide optimal biopsy, and radiation field mapping, with significant benefits for patient treatment. In addition, we did not find a significant association between RSI, diffusion metrics and MGMT promoter methylation.

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Conflicts of interest

Oslo University Hospital, Oslo, Norway, has received payment from NordicNeuroLab AS, Bergen, Norway for patents and/or intellectual property rights to author (K.E.E.).

Appendix A. Supplementary data

Supplementary material related to this article can be found, in the online version, at doi:https://doi.org/10.1016/j.ejrad.2020.109289.

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