ORIGINAL RESEARCH

Vessel Size Imaging is Associated with IDH Mutation and Patient Survival in Diffuse Lower-Grade Glioma

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Background: Patients with isocitrate dehydrogenase (IDH) mutant gliomas have better survival and appear to be more sensitive to chemotherapy than their IDH wild-type counterparts. We attempted to assess the correlations of vessel size imaging (VSI) values with IDH mutation status and patient survival in diffuse lower-grade glioma (LGG).

Methods: We enrolled 60 patients with diffuse LGGs, among which 43 had IDH-mutant tumors. All patients underwent VSI examination and VSI values for active tumors were calculated. Receiver operating characteristic (ROC) curves were established to evaluate the detection efficiency. Logistic regression was employed to determine the ability of variables to discriminate IDH mutational status. Kaplan–Meier survival analysis and Cox proportional hazards models were utilized to estimate the correlations of VSI values and other risk factors with patient survival.

Results: We observed that VSI values were lower in IDH-mutant LGGs than IDH wild-type LGGs. The VSImax and VSImean values had AUC values of 0.7305 and 0.7401, respectively, in distinguishing IDH-mutant LGGs from IDH wild-type LGGs. Logistic regression showed that VSImean values, age and tumor location were associated with IDH-mutant status, and the formula integrating the three factors had an AUC value of 0.7798 when distinguishing IDH-mutant LGGs from IDH wild-type LGGs. Moreover, LGG patients with high VSI values exhibited worse survival rates than those with low VSI values for both progression-free survival (PFS) and overall survival (OS). Multivariate Cox proportional hazards regression analysis suggested that IDH mutation status, VSImean values and multiple lesions or lobes were risk factors for PFS of LGG patients.

Conclusion: VSI value is associated with IDH genotype and maybe an independent predictor of the survival of patients with LGGs.

Keywords: vessel size imaging, glioma, isocitrate dehydrogenase, IDH, mutation, survival

Introduction

Infiltrating astrocytoma is the most common type of primary brain tumor and can be divided into grades II, III and IV according to the 2007 World Health Organization (WHO) classification.[1] Diffuse lower-grade glioma (LGG) is an infiltrative brain tumor consisting of WHO grade II and III neoplasms, which presents a more indolent course compared to glioblastoma (GBM, grade IV),[2,3] and LGG is also divided into a series of molecular subtypes based on isocitrate dehydrogenase (IDH) and 1p/19q co-deletion status according to the 2016 WHO classification.[1–3]

The prognosis of patients with glioma is related to several clinical factors, such as age, extent of tumor resection, and treatment modality,[4] and correlates with

molecular markers, such as IDH mutation, O6-methylguanine-DNA methyltransferase promotor methylation, and 1p/19q co-deletion. Notably, IDH mutation occurs in the majority of LGGs and has been considered a critical prognostic molecular marker for LGG.[4] Patients with IDH-mutant LGGs have better outcomes and appear to be more sensitive to chemoradiotherapy than those with IDH wild-type LGGs.[5,6] Therefore, identification of IDH genotype is beneficial for predicting the prognosis of LGG patients and for determining therapeutic strategies. Currently, however, determination of IDH genotype in LGGs requires invasive approaches, such as surgery or pathological biopsy, and then the use of an immunohistochemistry test, which may have an approximately 15% rate of missed diagnosis with the use of the antibody, R132H.[7] Therefore, it is urgently important to explore a noninvasive method for evaluating IDH mutation status.

Traditional magnetic resonance imaging (MRI) is a noninvasive imaging technology that generates anatomical images but cannot reflect tumor metabolism, so it is difficult to identify the molecular classification of gliomas. Functional MRI can obtain functional information by visualizing cortical activity and shows a certain effect in the molecular classification of gliomas, especially MR spectral imaging (MRS) and MR perfusion-weighted imaging (PWI).[8,9] For example, MRS can reach 80–90% accuracy in recognizing IDH mutation by detecting 2-hydroxyglutarate (2HG), a metabolite of IDH.[8] However, this technology is challenging and difficult to apply in clinical practice.[8] Cerebral blood volume (CBV) derived from PWI has been proposed to assess the prognosis of glioma patients and the molecular classification of glioma to some extent.[1,10] Vessel size imaging (VSI), another MR perfusion technology, can describe the structural heterogeneity of brain microvasculature based on a gradient-echo (GE)/spin-echo (SE) echo planar imaging sequence.[11] Our previous study indicated that VSI values are more accurate than relative CBV (rCBV) values in glioma grading,[12] suggesting that VSI may reflect the degree of malignancy. However, the efficiency of VSI in diagnosing the IDH genotype and predicting the prognosis of patients with LGGs has not been fully elucidated.

In the present study, we evaluated the diagnostic performance of VSI in recognizing IDH mutation status in LGGs and predicting the survival rates of LGG patients.

Materials and Methods
Patients

A total of 60 patients who underwent initial surgery and were histopathologically diagnosed with grade II or III glioma in Army Medical Center from March 2013 to May 2016 were included in this study. The characteristics of all patients are shown in Table 1. All tissue samples were tested for IDH mutation status. The inclusion criteria include the following: 1) the lesion is located in the supratentorial space; 2) the tumor did not spread, and the patient was over 18 years old; 3) chemotherapy was not performed before glioma surgery, and a complete treatment regimen and radiotherapy dose records were required for patients with recurrence; 4) IDH mutation and 1p/19q co-deletion status are known. Exclusion criteria include the following: 1) patient with other brain tumors, meningoencephalitis or tuberculosis; 2) pregnant women; 3) patient with mental disorders, uncontrolled diabetes or severe cardiopulmonary disorders. Approval for this retrospective study was obtained from the Ethics committee of Army Medical Center, and all patients have signed the informed consent. This study is an extension of a previously reported work.[12,13] This study was performed in accordance with the ethical standards of the 1964 Declaration of Helsinki and its most recent amendments.

Information on patient demographic characteristics and data on the extent of resection, adjuvant treatment, disease course, and survival were collected from our electronic medical records and through phone call follow-up. The extent of tumor resection was classified as total, subtotal (<100% and ≥75% of gross total removal), or partial (<75% of gross tumor removal) resection according to the integrative analysis of intraoperative impressions in surgery and postoperative MRI findings.[4] Multiple lobes and multiple lesions were defined as involvement of two or more lobes connected by direct routes of spread and multiple sites not connected by obvious routes of spread.[14] Progression-free survival (PFS) was defined as the time from diagnosis to tumor progression based on the Response Assessment in Neuro-Oncology (RANO) criteria[15,16] or the date of the last follow-up examination if the patient did not show disease progression. Overall survival (OS) was defined as the time from diagnosis to death or the date of the last follow-up examination if the patient did not die. Decisions on disease progression were made by the consensus of two neuroradiologists (H.-G and H.-Y.K., with 6 and 11 years of experience in brain MRI,

Table 1 The Clinical Characteristics of Patients

Characteristics/ Variables	IDH 1 Mutation Status		P value
	Mutated (n =43)	Wild (n =17)	
Age (mean ± SD, years)	41±10	50±17	0.059
Gender			
Male	22 (51.2%)	8 (47.06%)	1.000
Female	21 (48.8%)	9 (52.94%)	
Location			
Frontal lobe	17	2	NA
Temporal lobe	5	3	NA
Parietal lobe	1	1	NA
Occipital lobe	0	1	NA
Multiple lobes	18	10	NA
Multiple lesions	2	0	NA
WHO grade			
Grade II	30 (69.77%)	7 (41.18%)	0.075
Grade III	13 (30.23%)	10 (58.82%)	
VSImax (mean ± SD, μm)	106.45 ±60.3	155.7±55.61	0.005
VSImean (mean ± SD, μm)	74.06±44.7	113.41±44.42	0.003
Extent of resection			
Gross total resection	25	9	NA
Subtotal resection	8	5	NA
Partial resection	10	3	NA
Histopathology			
Diffuse astrocytoma	12	4	NA
Oligodendroglioma	11	3	NA
Oligoastrocytoma	7	0	NA
Anaplastic astrocytoma	1	6	NA
Anaplastic oligodendroglioma	7	2	NA
Anaplastic oligoastrocytoma	5	2	NA
1p/19q			
co-deletion	28 (65.12%)	5 (29.41%)	0.020
intact	15 (34.88%)	12 (70.59%)	
Adjuvant therapy			
Radiation	4	1	NA
Chemotherapy	9	4	NA
Chemoradiotherapy	21	9	NA
Untreated	3	3	NA
Unknown	6	0	NA

Notes: Except where indicated, data are numbers of patients, and numbers in parentheses are percentage. Independent sample *t*-tests or Mann–Whitney *U*-tests, as appropriate, were used to compare the differences in continuous variables. The Chi-square testing was used to analyze the constituent ratio of variables. NA, not applicable.

respectively) or based on the pathological diagnosis (if available). The data of patients without an event were censored at the date of the most recent follow-up, regardless of whether they were scheduled for future follow-ups or had been lost to follow-up.

IDH Mutation and 1p/19q Co-Deletion Detection

IDH mutation status was determined using polymerase chain reaction (PCR) and pyrosequencing. Detailed protocols of these methods have been described elsewhere.[17] Briefly, genomic DNAs were isolated from paraffin-embedded tumor tissues with a QIAamp DNA Mini Kit (Qiagen) according to the manufacturer's protocol. Forward primer 5′-CGGTCTTCAGAGAAGCCATT-3′ and reverse primer 5′-GCAAAATCACATTATTGCCAAC-3′ for IDH1 were used for PCR amplification. Fluorescence in situ hybridization (FISH) was used to assess 1p/19q co-deletion.

MRI Protocol

Preoperative MRI was performed using a 3T scanner (Magnetom Verio; Siemens, Erlangen, Germany). The routine imaging protocol included the following sequences: sagittal T1-weighted spin-echo (TR/TE, 440ms/2.48ms), axial T2-weighted fast spin-echo (FSE) (TR/TE, 4900ms/96ms), axial T1-weighted spin-echo sequence (TR 250ms, TE 2.67ms) performed before and after intravenous administration of gadolinium-DTPA (Magnevist, Bayer-Schering), 5mm slice thickness, 1mm intersection gap. MRI data for vessel size imaging (VSI) was acquired using a 1.5T system (GE Healthcare) with following sequences: i.v. injection of Gd-DTPA using a gradient-echo spin-echo (GE-SE) sequence, with acquisition parameters: field of view = 24 × 24cm, TR 1500ms, TE (GE) 30ms, TE (SE) 100ms, flip angles = 90°, matrix size = 64 × 64, NEX 1, slice number was 7, and 50 images were obtained at each slice. As well, 0.2mmol of gadobutrol per kilogram of body weight was injected at a rate of 3mL/s and immediately followed by a 20mL bolus of saline.

VSI Measurement

VSI images were analysed using an Advantage Workstation (version 4.9; General Electric Medical Systems) equipped with a dedicated software package (VSI; GE Medical Systems). Detailed protocols for the post-acquisition image processing have been described elsewhere.[11] The VSI image threshold was adjusted to

0–80μm or 0–120μm. The MR T2-weighted original image was used as a guide for tumor location. A hand-drawn ROI corresponding to the hot-spot area avoiding any blood vessels and necrotic parts in the tumor tissue on the VSI map was chosen for the calculation of VSI values. Circular ROIs of 50–60mm^2 were placed on the tumoral parenchyma to yield quantitative data, and the circular ROIs were obtained from at least five adjacent regions. The average and max of the obtained values were named the VSImean values and VSImax values, respectively. Two experienced neuroradiologists (H.-Y.K. and H.-G, with 11 and 6 years of experience, respectively) were blinded to the histopathologic diagnosis and completed measurements, independently.

Immunohistology Analysis

An immunohistology kit (Zhong Shan-Golden Bridge Biological Technology company, Beijing, China) was used to detect CD34 expression in the tumor tissues embedded using paraffins according to the manufacturer's instruction. The results were observed using a microscope (Olympus BX41, France) and then analysed using CellSens Standard software. The microvascular diameter was calculated from at least five fields of the largest diameter using Image-Pro Plus 5.0 software. The measurements were performed by an experienced pathologist (H.-L.X, with>20 years of experience) who was blinded to the MRI results.

Statistical Analysis

All results are expressed as the mean ± SD or median ± 10–90 percentile. The *P* value of less than 0.05 is considered statistically significant. Inter-observer reliability of VSI value measurements between the two radiologists was evaluated by intraclass correlation coefficients (ICC); ICC > 0.75 indicated excellent agreement. Independent-samples *T*-test or Mann–Whitney *U*-test was used to analyse the differences in VSI values between patients with IDH-mutant glioma and those with IDH wild-type glioma and between patients with grade II glioma and those with grade III glioma. The Chi-square testing was used to analyze the constituent ratio of variables. Pearson correlation analysis was used to evaluate the relationship between VSI values and histological microvascular diameters. A receiver operating characteristic (ROC) curve was used to assess the specificity and sensitivity of VSImean and VSImax in distinguishing IDH-mutated LGGs from IDH wild-type LGGs, and the optimal cutoff values were determined using the Youden index. Logistic regression was used to determine the ability of variables to discriminate IDH mutational status. The PFS and OS (4 years) of patients were analysed using Kaplan–Meier curves and the Log-rank test. For PFS, Cox regression analysis was employed to assess the risk factors, including gender, age, tumor location, adjuvant therapy, extent of resection, histopathology type, IDH mutation status, 1p/19q co-deletion and VSI values. GraphPad Prism 8.0.1 and R (version 3.6.2) software were used for statistical analysis.

Result
VSI Values Correlate with Microvessel Diameters in LGGs

There was an excellent interobserver agreement for the VSImean (ICC = 0.955) and VSImax (ICC = 0.923) measurements, and the VSImax and VSImean values were calculated based on the average values of the two observers. To investigate whether VSI values are associated with microvessel diameters in LGGs, we obtained VSImax and VSImean values and microvessel diameters in 60 LGG samples. We found that both VSImax and VSImean values significantly correlated with microvessel diameters in LGGs (Figure 1A and B), suggesting that VSI and histopathological approaches have a high consistency in evaluating the microvessel size in LGGs.

VSI Values are Lower in IDH-Mutant LGGs and in Grade II Gliomas

Next, we sought to analyse the association of VSI values with IDH mutation status and glioma grading. We found that both VSImax and VSImean values were significantly lower in IDH-mutant LGGs than in IDH wild-type LGGs (Figure 2A and E). Moreover, the VSImax and VSImean values were also significantly lower in grade II gliomas with IDH mutation than in those with IDH wild type (Figure 2B and F), whereas in grade III gliomas, there were no significant differences between IDH mutation and IDH wild type (Figure 2C and G). Furthermore, we also found that the VSImax and VSImean values were significantly lower in grade II gliomas than in grade III gliomas (Figure 2D and H). The representative images of MRI and pathology are shown in Figure 3. These results suggest that the VSImax and VSImean values correlate with IDH mutation status and glioma grading.
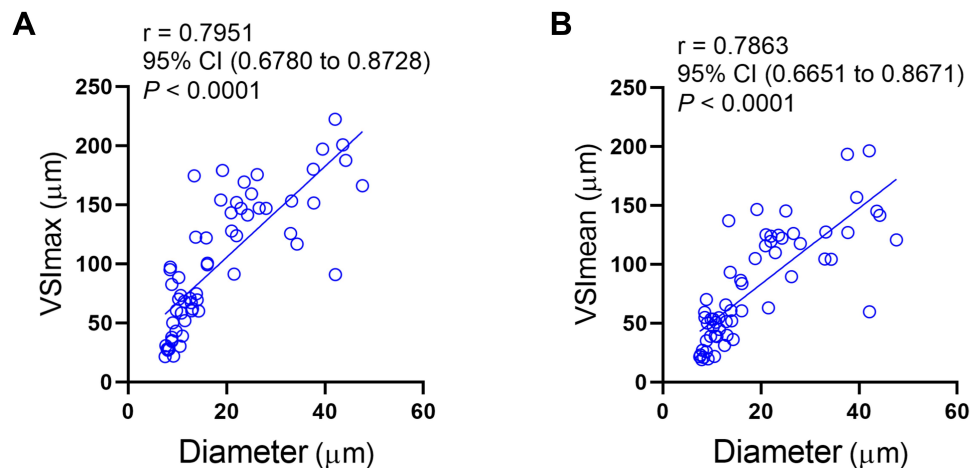
Figure 1 The VSI values correlate with microvessel diameter in LGGs. (**A**) VSImax and (**B**) VSImean values positively correlate with microvessel diameter in 60 LGG patients.
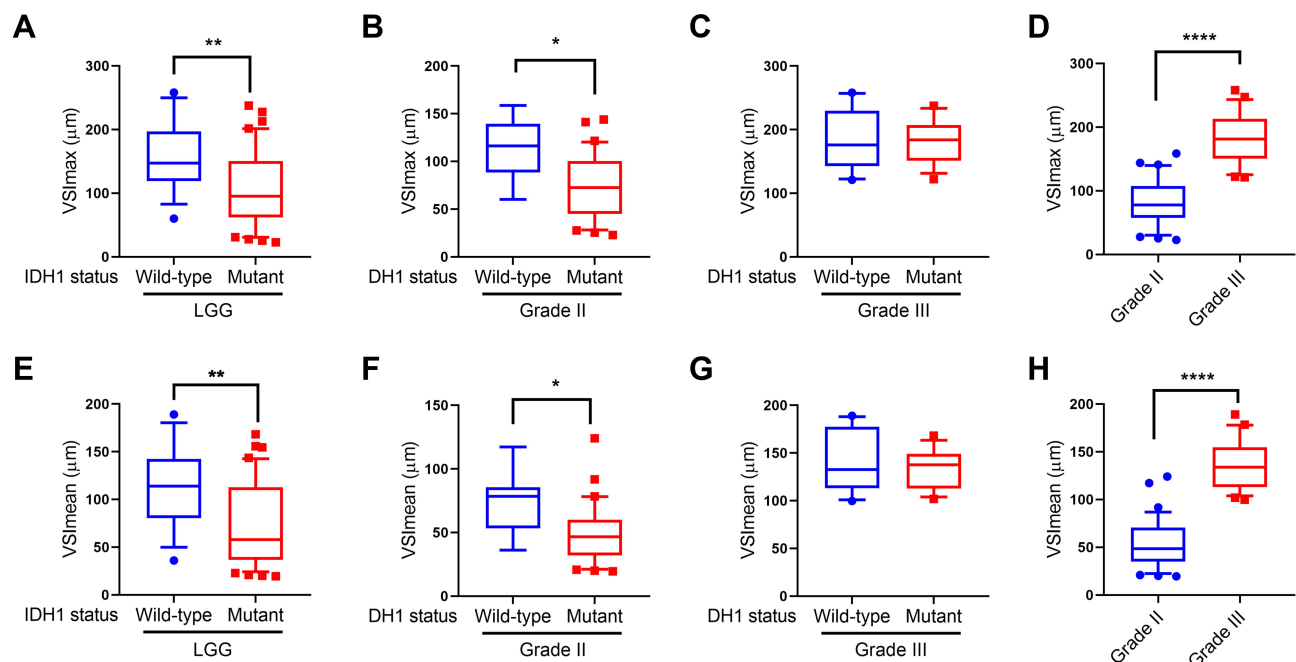

Figure 2 The VSI values are lower in IDH-mutant LGGs and in grade II gliomas. (**A** and **E**) IDH wild-type LGGs (n = 17) and IDH-mutant LGGs (n = 43). (**B** and **F**) Grade II gliomas with IDH wild type (n = 7) and grade II gliomas with IDH mutation (n = 20). (**C** and **G**) Grade III gliomas with IDH wild type (n = 10) and grade III gliomas with IDH mutation (n = 13). (**D** and **H**) Grade II gliomas (n = 37) and grade III gliomas (n = 23). Boxes show the VSI values (μm) of different types of LGGs, with appearance: box and whiskers, and whiskers: 10–90 percentile. *P < 0.05; **P < 0.01; ****P < 0.0001.

VSI Values May Serve as a Marker for the Prediction of IDH Mutation Status in LGGs

Then, we analysed the potential power of the VSImax and VSImean values as markers in differentiating IDH-mutant LGGs from IDH wild-type LGGs. ROC curve analyses revealed that VSImax had an AUC value of 0.7305 in distinguishing IDH-mutant LGGs from IDH wild-type LGGs, with 62.79% sensitivity and 82.35% specificity

when an optimal cutoff value of 112.8 μm was used (Figure 4A), while VSImean had an AUC value of 0.7401 with 65.12% sensitivity and 82.35% specificity when an optimal cutoff value of 78.5 μm was used (Figure 4B). We sought to combine the VSImean values with other factors including the VSImax values, age and tumor location using a stepwise logistic regression method, and a formula (IDH-mutant signature = −5.42125 + VSImean × 0.01550 + tumor location ×
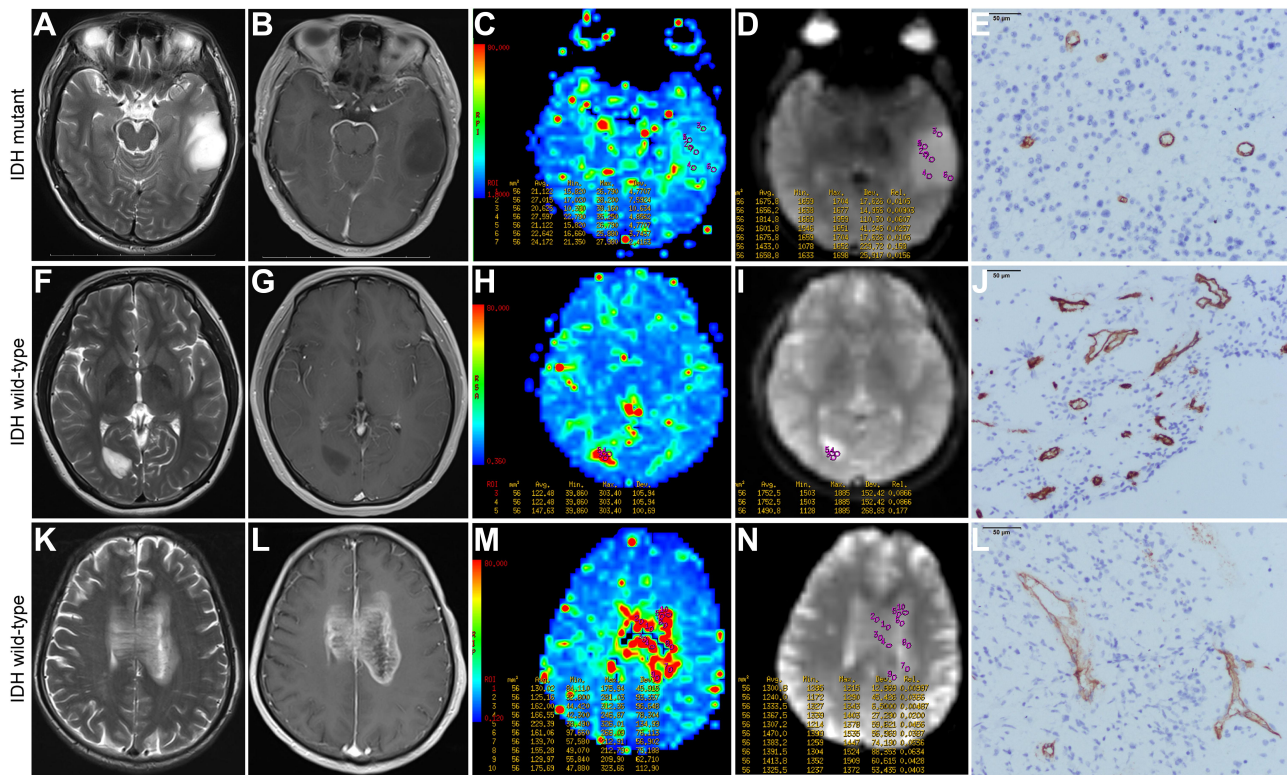
Figure 3 MRI, VSI map and CD34 staining of IDH-mutant and wild-type LGGs. A 31-year-old man with an IDH-mutant diffuse astrocytoma (WHO grade II) at the left temporal lobe showing homogeneous high signal intensity on a T2-weighted image (**A**) and no enhancement on a contrast-enhanced T1-weighted image (**B**). ROIs were drawn on the area showing the highest signal within the lesion on the processed VSI map image (**C**) and a corresponding raw VSI image (**D**). Representative photograph of CD34 staining for the corresponding tumor micro-vessel (**E**). The VSImean is 23.47μm and VSImax is 27.597μm, which belongs to the VSI-low group according to the optimized cutoff values for OS and PFS prediction. The PFS of this patient was more than 60 months. This patient was alive at the last follow-up. A 19-year-old woman with an IDH wild-type diffuse astrocytoma (WHO grade II) at the right occipital lobe showing similar image features on a T2-weighted image (**F**) and a contrast-enhanced T1-weighted image (**G**) as the IDH-mutant diffuse astrocytoma case. ROIs were drawn on the area that showed the highest signal on the processed VSI map image (**H**) and a raw VSI image (**I**). Representative photograph of CD34 staining for the corresponding tumor micro-vessel (**J**). The VSImax value is 147.63 μm and the VSImean value is 82.26μm, which belongs to the VSI-high group according to the optimized cutoff values for OS and PFS prediction and is close to cutoff values for OS and PFS prediction. The PFS of this patient was more than 46 months. They both underwent gross total resection and chemoradiotherapy after surgery. A 72-year-old man with an IDH wild-type anaplastic astrocytoma (WHO grade III) at the body of the corpus callosum and cingulate gyrus showing heterogeneous high signal intensity on a T2-weighted image (**K**) and intense but inhomogeneous enhancement on a contrast-enhanced T1-weighted image (**L**). ROIs were drawn on the area that showed the highest signal on the processed VSI map image (**M**) and a raw VSI image (**N**). Representative photograph of CD34 staining for the corresponding tumor micro-vessel (**L**). The VSImax value is 229.39μm and VSImean value is 156.7μm, which belongs to the VSI-high group according to the optimized cutoff values for OS and PFS prediction. He underwent a subtotal resection and without any therapy after surgery. The OS of this patient was only 12 months.
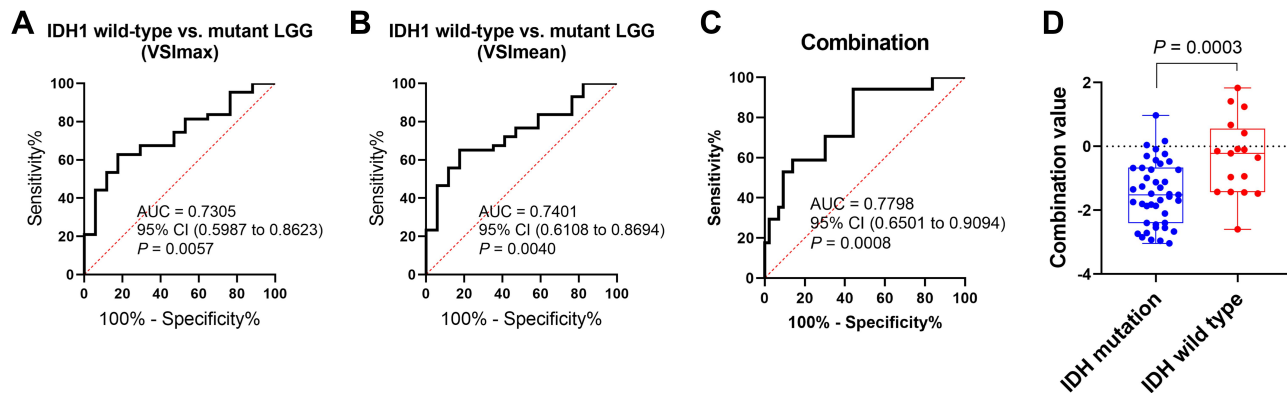

Figure 4 VSI values may serve as a marker for the prediction of IDH mutation status in LGGs. ROC curves were established. IDH wild-type LGG cases (n = 17) versus those IDH-mutated LGG cases (n = 43). (**A**) ROC curve: VSI max. (**B**) ROC curve: VSImean. (**C**) Combination of the VSImean values with tumor location and age using stepwise logistic regression. (**D**) Combination value between IDH-mutant LGGs (n = 43) and IDH wild-type LGGs (n = 17).

0.85926 + age × 0.03618) was obtained. Then, we used this formula to integrate the three factors and obtained an AUC value of 0.7798 to distinguish IDH mutation and IDH wild type, with 94.12% sensitivity and 55.81% specificity (Figure 4C and D). However, the VSImax values were not included in this formula, possibly due to their insufficient effect. These results suggest that the VSImean values and integrated factors have the potential power to predict IDH mutation status in LGGs.

VSI Values are Associated with the PFS and OS of Patients with LGGs

Next, we sought to analyse whether the VSI values correlate with the PFS and OS of patients with LGGs. The follow-ups of 60 patients with LGGs include: 32 of 60 patients had tumor progression (10 of 32 patients with IDH wild-type LGGs and 22 of 32 patients with IDH-mutant LGGs); 16 patients died (7 of 17 patients with IDH wild-type LGGs and 9 of 47 patients with IDH-mutant LGGs); and 9 of 60 patients were lost to follow-up. The median PFS of the patients was 42.0 months. As the survival probability did not fall below 0.5, the median OS was not calculated. The mean OS of the patients was 61.279 months. We then divided the patients into different groups for analysis at four-year follow-up based on corresponding cutoff values (VSImax-Low versus VSImax-High and VSImean-Low versus VSImean-High), IDH mutation status (IDH wild-type versus IDH mutant) and glioma grading (grade II versus grade III). The mean OS of the VSImax-High group (36.515 months; 95% CI, 30.415–42.615) and VSImean-High group (35.970 months; 95% CI, 29.683–42.258) was significantly shorter than the VSImax-Low group (44.464 months; 95% CI, 40.563–48.365) and VSImean-Low group (44.604 months; 95% CI, 40.847–48.362) (Figure 5A and B). And the mean PFS of the VSImax-High group (27.437 months; 95% CI, 20.936–33.938) and VSImean-High group (26.707 months; 95% CI, 20.089–33.325) was significantly shorter than the VSImax-Low (38.962 months; 95% CI, 33.655–44.268) and VSImean-Low groups (39.178 months; 95% CI, 34.053–44.303) (Figure 5C and D). The representative images are shown in Figure 3. The mean OS and PFS of the IDH wild-type group were also significantly shorter than that of the IDH-mutant group (Figure S1A and D). In addition, the mean OS and median PFS of the grade III group were both shorter than those of the grade II group (Figure S1B and E), but they were not significantly

different between IDH wild-type grade II and grade III groups (Figure S1C and F).

IDH Mutation Status, VSImean, and Multiple Lesions or Lobes are Risk Factors for PFS in Patients with LGGs

Next, we sought to analyse the risk factors including the VSI values, IDH status, 1p/19q co-deletion status and multiple clinical characteristics for PFS in patients with LGGs. By performing univariate Cox analysis, we found that IDH mutation status, VSImax values, VSImean values, grade, age, adjuvant therapy, and multiple lesions or lobes were associated with PFS, but gender, histopathology type, 1p/19q co-deletion status and extent of resection were not correlated with PFS (Table 2). Furthermore, according to the results of univariate analysis, four models for predicting PFS were generated using multivariate Cox regression analysis. We found that the factors of multiple lesions or lobes, IDH mutation status and VSImean values were hazard factors for PFS in different models (Table 3).

Discussion

In this study, we found that the VSImax and VSImean values are lower in IDH-mutant LGGs than in IDH wild-type LGGs and in grade II gliomas than in grade III gliomas; ROC curve analyses suggested that VSI values may serve as a potential marker for distinguishing IDH-mutant LGGs from IDH wild-type LGGs; and the combination of VSImean values, age and tumor location showed more power to predict IDH genotype in LGGs. Furthermore, we demonstrated that LGG patients with high VSI values have worse survival rates than those with low VSI values at the four-year follow-up for PFS and OS. Moreover, univariate Cox analysis revealed that VSI values, IDH mutation status, grade, age, adjuvant therapy, and multiple lesions or lobes were risk factors for the survival of LGG patients. Multivariate Cox proportional hazards regression analysis suggested that IDH mutation status, VSImean values and multiple lesions or lobes might be independent risk factors for PFS of LGG patients. These suggested that the VSImean values may be potential imaging features that can be used to detect IDH mutation status and patient survival in LGGs.

There are differences in angiogenesis between IDH wild-type and mutant gliomas.[1,10] Studies have indicated that IDH wild-type LGGs exhibit high enrichment of
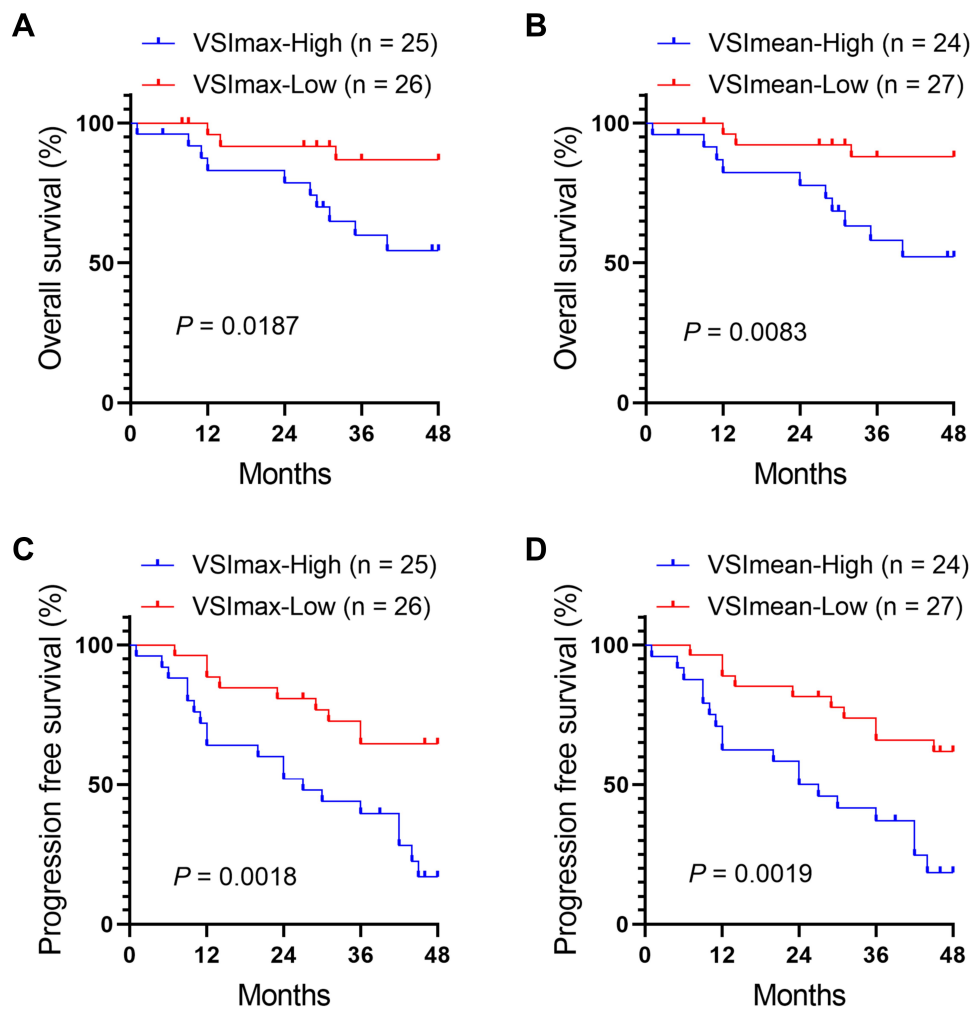
Figure 5 VSI values are associated with the PFS and OS of patients with LGGs at the four-year follow-up. Kaplan–Meier curve survival analyses show the OS and PFS of patients with different types of LGGs. (**A** and **C**) The patients were divided into two groups of VSImax-High and VSImax-Low, based on a VSImax cutoff value of 112.8μm; (**B** and **D**) the patients were divided into two groups of VSImean-High and VSImean-Low, based on VSImean cutoff value of 78.5μm.

anoxia- and angiogenesis-associated genes and an increased pericyte coverage of micro-vessels compared with IDH-mutant LGGs.[18,19] The variation of glioma angiogenesis can be differentiated using a dynamic susceptibility contrast (DSC) perfusion MRI model,[1,10] where rCBV is a representative parameter that is widely used.[1,20,21] However, another relatively new parameter, VSI, has rarely been reported. VSI can describe the structural heterogeneity of the brain microvasculature as well as the microvascular oxygen content and spatial arrangement.[11] We also found that VSI values are positively correlated with microvessel diameter. VSI was calculated based on curves of the GE relaxation rate R2* and SE relaxation rate R2 in the signal drop portion.[22] Choi et al found that using representative T2* susceptibility signal intensity–time curves from DSC perfusion MRI to distinguish the IDH genotype will obtain a better effect than rCBV because these curves have

different upslopes in the signal drop portion.[7] In this study, our findings revealed that VSI values act as a marker to distinguish IDH wild-type LGGs from IDH-mutant LGGs, suggesting that VSI may be an alternative method for the evaluation of IDH mutation status in LGGs. Our previous study indicated that VSI is more accurate than rCBV in grading glioma.[12] However, we did not compare the effect of VSI with rCBV on the evaluation of IDH mutation status in the present study; therefore, it is necessary to evaluate which one is better in the future. In addition, using stepwise logistic regression, we found that the VSImean values, age and multiple lesions or lobes were independent variables for identifying the IDH genotype, and their combination had a higher AUC. In line with our study, Villanueva-Meyer et al also showed multifocal tumor and age were independently associated with IDH mutation status.[14]

Table 2 Univariate Survival Analysis

Features	Hazard Ratio	95% Confidential Interval	P value
VSImean	1.011	1.004, 1.019	0.0029
VSImax	1.009	1.003, 1.015	0.0033
IDH mutation	0.3103	0.1409, 0.6836	0.0037
Grade	3.073	1.432, 6.593	0.0040
Multiple lesions or lobes	2.659	1.364, 5.184	0.0041
Age	1.036	1.004, 1.068	0.0254
Adjuvant therapy after surgery	0.6957	0.4847, 0.9987	0.0492
Gender	1.32	0.6563, 2.653	0.4360
1p/19q co-deletion	0.7687	0.3812, 1.55	0.4620
Histopathology	0.8642	0.5708, 1.308	0.4900
Extent of resection	1.031	0.6732, 1.579	0.8890

It is well known that patients with IDH-mutant LGGs have better outcomes than those with IDH wild-type LGGs,[6–8] and grade III glioma leads to poorer survival rates compared with grade II glioma, but the survival time is not significantly different between grade II and grade III IDH wild-type gliomas.[5] Our study also confirmed the same results using Kaplan–Meier survival analysis and Cox proportional hazards models, and we found that IDH mutation status rather than grade was an independent prognostic factor for PFS. Interestingly, we are the first to observe that LGG patients with high pre-treatment VSI values had worse survival rates, both PFS and OS. A few studies have shown that vascular normalization can improve tumor perfusion and thereby increase oxygenation, which can improve the OS of patients with glioblastoma, and these patients with vascular normalization after cediranib, which can be evaluated by VSI, also have longer OS and PFS.[17,23] Our findings suggest that the pre-treatment VSI value is a predictive factor for the outcome of LGG patients, and it does not depend on the treatment strategy or vessel change after treatment. Furthermore, we found that VSImean values had a better effect than VSImax values in predicting LGG patient outcomes. The underlying reason may be that the VSImean values were built from multiple values in hot-spot areas, so they can reflect the integrated status of glioma, but the VSImax values were obtained from the highest value, which more easily results in a bias.

Our analyses of univariate Cox regression indicated that, except for the VSI values and IDH mutation status, several clinical features were correlated with LGG patient outcomes, including older age, postoperative adjuvant chemoradiotherapy, and multiple lesions or lobes, and a large number of studies support our findings.[24–26] For multivariate Cox regression, we established four models based on different risk factors obtained from the results of univariate analysis. Each model contains three factors at most, due to the limit on the number of our cases. We found that the VSImean values rather than the VSImax values may be an independent risk factor for LGG patients. We further found that the VSImean values and IDH mutation status are two independent risk factors when we combine each of the two factors with two other clinical factors. These results suggested that when VSI serves as a risk factor, its efficiency is similar to that of IDH for predicting LGG patient outcomes. Previous studies have indicated that the pre-treatment rCBV value is also an independent prognostic factor of glioma, and decreased rCBV values correlate with longer OS.[21,27–29] VSI and rCBV were two different parameters in perfusion imaging that reflect different hemodynamic changes and microvascular structures, and their comparison with regard to the OS or PFS of patients with LGGs needs to be explored in the future. However, factors including extent of resection, 1p/19q co-deletion status and histopathology type were not associated with PFS in our study, although these factors may be prognostic factors for glioma.[30,31] Several studies obtained the same results as our study. For instance, Bio Joo et al indicated

Table 3 Multivariate Prognostic Models, Clinical Factors with IDH or VSI Values, for Predicting PFS

Features	Model 1		Model 2		Model 3		Model 4	
	HR(95% CI)	P value	HR(95% CI)	P value	HR(95% CI)	P value	HR(95% CI)	P value
IDH	0.4103 (0.1658, 1.015)	0.05397			0.3548 (0.1552, 0.8111)	0.0140		
VSImean			1.0013 (0.9864, 1.017)	0.85896			1.0095 (1.0015, 1.018)	0.0202
Grade	2.1883 (0.9146, 5.235)	0.07850	2.7060 (0.5777, 12.674)	0.20638				
ML	2.8187 (1.3947, 5.697)	0.00389	2.6494 (1.2996, 5.401)	0.00734				
Age					1.0243 (0.9954, 1.542)	0.1006	1.0220 (0.9929, 1.052)	0.1401
Therapy					0.7156 (0.4944, 1.0358)	0.0761	0.7523 (0.5264, 1.075)	0.1185

Notes: HR, hazard ratio; 95% CI, 95% confidential interval; IDH, IDH mutation; ML, multiple lesions or lobes; Therapy, adjuvant therapy after surgery.

that extent of resection was not related to PFS and OS in high-grade glioma;[4] another study showed that there was no difference in either PFS or OS among the various histologies in WHO grade II gliomas;[32] Villanueva-Meyer et al indicated that the 1p/19q co-deletion is associated with the prognosis of patients with grade II gliomas, but there were no IDH wild-type tumors with 1p/19q co-deletion among their cases.[14] Different from those cases, our patients have IDH wild-type LGG with 1p/19q co-deletion, and Hameed et al revealed that 1p/19q co-deletion was not a significant predictor of OS in LGGs.[33]

Nevertheless, our study has several limitations. First, we used retrospective data, our sample size was relatively small. Due to the loss of follow-up for some patients, there were a lot of censored data, which affected the prognosis analysis, especially the accuracy of the judgment of overall survival, and most patients with IDH-mutant LGGs were still alive when the follow-up had been finished, so we did not conduct Cox regression analysis of OS. Second, we manually measured the VSI values, which will be influenced by some subjective factors, but because we used the same threshold to observe the imaging and the VSI values are absolute parameters, there was high consistency between different observers, suggesting that this method has acceptable repeatability. A previous study indicated that there was a slightly better correlation of Automatable Gaussian Normalized CBV with the OS of glioma patients than standard white matter normalized CBV, but the difference was not statistically significant.[31] Therefore, we still obtain a good conclusion by using the hot-spot method, but more objective measurement methods still need further study.

In conclusion, our findings indicated that VSI values may serve as a predictor of IDH genotype in LGGs and of the prognosis of LGG patients.

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Disclosure

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