ORIGINAL REPORT

The heterogeneity of blood flow on magnetic resonance imaging: A biomarker for grading cerebral astrocytomas

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KEYWORDS
Astrocytoma;
Magnetic resonance;
Perfusion;
Brain;
Brain tumors;
Kurtosis

Abstract
Objectives: To study whether the histograms of quantitative parameters of perfusion in MRI obtained from tumor volume and peritumor volume make it possible to grade astrocytomas in vivo.

Materials and methods: We included 61 patients with histological diagnoses of grade II, III, or IV astrocytomas who underwent T2*-weighted perfusion MRI after intravenous contrast agent injection. We manually selected the tumor volume and peritumor volume and quantified the following perfusion parameters on a voxel-by-voxel basis: blood volume (BV), blood flow (BF), mean transit time (TTM), transfer constant (Ktrans), washout coefficient, interstitial volume, and vascular volume.

For each volume, we obtained the corresponding histogram with its mean, standard deviation, and kurtosis (using the standard deviation and kurtosis as measures of heterogeneity) and we compared the differences in each parameter between different grades of tumor. We also calculated the mean and standard deviation of the highest 10% of values. Finally, we performed a multiparametric discriminant analysis to improve the classification.

Results: For tumor volume, we found statistically significant differences among the three grades of tumor for the means and standard deviations of BV, BF, and Ktrans, both for the entire distribution and for the highest 10% of values. For the peritumor volume, we found no significant differences for any parameters. The discriminant analysis improved the classification slightly.

Conclusions: The quantification of the volume parameters of the entire region of the tumor with BV, BF, and Ktrans is useful for grading astrocytomas. The heterogeneity represented by the


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La heterogeneidad del flujo sanguíneo en resonancia magnética, biomarcador para clasificar por grados los astrocitomas cerebrales

Resumen

Objetivos: Estudiar si los histogramas de los parámetros cuantitativos de perfusión por RM obtenidos a partir de los volúmenes tumoral y peritumoral permiten clasificar in vivo el grado de los astrocitomas.

Material y métodos: Se incluyen 61 pacientes diagnosticados histológicamente de astrocitoma grado II, III o IV, estudiados mediante RM de perfusión T2* con contraste intravenoso, seleccionando manualmente los volúmenes tumoral y peritumoral, cuantificándose voxel a voxel diferentes parámetros de perfusión: volumen sanguíneo (VS), flujo sanguíneo (FS), tiempo de tránsito medio (TTM), constante de transferencia ($K_{trans}$), coeficiente de lavado, volumen intersticial y volumen vascular.

Para cada volumen se obtuvo el histograma correspondiente con su media, desviación típica y curtosis, estas últimas como medidas de heterogeneidad, comparándose las diferencias por parámetro y grado tumoral. También se calcularon la media y desviación del 10% de los valores máximos. Finalmente se realizó un análisis discriminante multiparamétrico para mejorar la clasificación.

Resultados: En el volumen tumoral se obtuvieron diferencias estadísticamente significativas entre los 3 grados tumoriales para la media y la desviación de VS, FS y $K_{trans}$, tanto para la distribución completa, como para el 10% máximo. En la región peritumoral no se obtuvieron diferencias significativas para ningún parámetro. El análisis discriminante mejoró ligeramente la clasificación.

Conclusiones: La cuantificación de parámetros del volumen total de la región tumoral con VS, FS y $K_{trans}$ es útil para establecer el grado de los astrocitomas. La heterogeneidad, representada por la desviación típica del FS, es el parámetro con mayor fiabilidad diagnóstica para separar los tumores de bajo y alto grado.

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Introduction

From the histological viewpoint astrocytomas are heterogeneous neoplasms in which in the same tumor both low- and high-grade areas coexist the latter being the ones that define the true histological differentiation. Vascular proliferation is one of the histopathological descriptors used to categorize glial tumors. These neoformed vessels show anomalies during their development, maturation and distribution within the neoplastic tissue and the surrounding peritumoral region. On the other hand they have an infiltrating nature enabling the coexistence of both healthy and tumor tissues in the surrounding brain tissue. This is why with the help of perfusion studies using magnetic resonance images (MRI) and spectroscopy with MRI the characteristics of peritumoral area have been used to distinguish between glial and metastatic lesions.

Perfusion studies using MRI give us information on how angiogenesis and patency can alter the tumor vessels. As a matter of fact the parameters derived from the quantitative analysis of perfusion help us categorize tumor degrees in a much better way than conventional MRI-based on morphological criteria only.

The quantitative analysis of perfusion uses a monocompartmental mathematical model assuming that the contrast media remains in the intravascular space and is not extravasated into the interstitial media. From that model the tissue values of blood volume (BV), mean transit time (MTT) and blood flow (BF) can be obtained usually as a measure on the healthy white matter. Since tumors are dysfunctional when it comes to the patency of the hematoencephalic barrier (HEB) with extravasation of the contrast media from the vascular to the interstitial compartment, a 2-compartment model is used to measure the transfer dynamics of contrast media by using the transfer coefficient ($K_{trans}$), the vascular ($v_v$) and interstitial volumes ($v_e$) and the cleanser coefficient ($k_{ep}$). Several studies have shown that there is a good correlation between the BV and the tumor stage of astrocytomas which allows us to distinguish between medium-grade (II) and high-grade astrocytomas (III and IV). The relation between $K_{trans}$ and the tumor stage is more controversial. Even though the data from perfusion studies using MRI are usually analyzed using average values from the regions of interest (ROI) a more accurate way to evaluate changes is to analyze the histogram of ROI or of the whole tumor
volume. Histogram offers a graphic representation of the frequencies of appearance of the regional values reached for every variable allowing us to analyze the distribution of each parameter in each and everyone of the regions analyzed.

Our hypothesis is that tumor degree in astrocytomas influences the perfusion parameters with MRI both in the tumor and peritumor areas. The goal of this work is to study the feasibility of quantitative parameters of perfusion in both the monocompartmental and pharmacokinetic models analyzed in the whole tumor and peritumor volumes through the analysis of histograms to categorize histological degrees in a large series of astrocytomas.

Materials and methods

Subjects

We did a retrospective study by reviewing the clinical history and the brain perfusion studies using MRI of patients presented consecutively in the Neuro-oncology Committee of our center between January 2006 and June 2011 with histological diagnosis of astrocytoma located in the supratentorial region. There were no pediatric patients as they did not go through this committee since they are always referred, as there is no Oncology or Neurosurgery hospital for children. Oligodendroglial tumors were not included in the filtering of the Neuro-oncology Committee database since a significant number of low-grade odendrogliomas can have a high BF that is missing in the histopathology. This is how we got 113 patients with diagnostic confirmation and perfusion studies using MRI.

Exclusion criteria for the selection of patients were also taken into consideration (1) perfusion studies using MRI of patients whose data could not be collected (n = 51) due to movement artifacts and lack of collaboration that is when patients’ exacerbated movements coincided with the acquisition of volumes of signal loss caused by IV contrast (1st step) not allowing us to quantify perfusion parameters; (2) patients diagnosed with grade I-pilocytic astrocytomas since this kind of tumors have different characteristics showing one tumor nodule enhanced after IV contrast and often showing BF and high patency categorizing them in a different group of lesions when it comes to the characteristics of perfusion using MRI (n = 1).

Finally there were 61 patients (44 males and 16 women) with supratentorial astrocytomas with histopathological confirmation based on the classification of the World Health Organization. The material was obtained after surgical resection in 36 patients (59%) or through neuronavigational-guided biopsy in 25 patients (41%). The distribution according to grades was 10 patients with grade II-tumors (16%) of which 8 were infiltrating astrocytomas and 2 oligoastrocytomas; 12 grade III-tumors or anaplastic astrocytomas (20%); and 39 grade IV-tumors (glioblastomas) (64%).

All patients underwent a perfusion study using MRI with the dynamic contrast material-enhanced T2*-weighted perfusion MRI after the administration of a paramagnetic contrast media agent. No patient showed impaired renal function that would counter indicate the administration of IV contrast (gadodiamide). The studies were done before the administration of any oncological therapies including the used corticoids yet this was not a compulsory stipulation for the selection of patients.

The average age of patients was between 26 and 74 years (55.8 ± 13.6 years). The location of tumors was: 27 tumors located in temporal lobes (44.3%); 20 frontal ones (32.8%); 9 were parietal (14.8%); one was located in the occipital lobe (1.6%); one was a thalamic tumor (1.6%) and 3 were tumors with affection of 2 adjacent lobes (4.9%).

Consent from the hospital Ethical Committee was obtained for the work and further publication of this report yet the MRI studies of patients were obtained within the usual medical practice. For the analysis of images all personal information on the patients was eliminated.

Image acquisition

MRI explorations were done through 1.5T imaging equipment (Philips Intera®, Philips Healthcare, The Netherlands) with an 8-channel multiple-element head coil. All patients underwent a conventional study included these sequences:

- Sagittal TSE-T1 (TR 500 ms, TE 20 ms, voxel size 0.5 mm × 0.5 mm × 5 mm).
- Coronal TSE-FLAIR (TR 1.100 ms, TE 140 ms, TI 2800 ms, voxel size 0.5 mm × 0.5 mm × 6 mm).
- Transverse TSE-T2 (TR 2.000 ms, TE 120 ms, voxel size 0.4 mm × 0.4 mm × 5 mm).
- Transverse TSE-T1 (TR 500 ms, TE 20 ms, voxel size 0.4 mm × 0.4 mm × 5 mm).
- Enhanced Transverse Diffusion (TR 2.946 ms, TE 74 ms, b values of 0 and 1.000 s/mm², voxel size 0.9 mm × 0.9 mm × 5 mm).
- IV Contrast TSE-T1 Sequences (TR 500 ms, TE 20 ms, voxel size 0.5 mm × 0.5 mm × 5 mm) acquired in 3 planes after perfusion study.

The perfusion study was acquired through the dynamic contrast material-enhanced T2*-weighted perfusion MRI. Echo-planar imaging (EPI) was done with segmentation of gradient-echo (GRE) with a TR 836 ms, TE 30 ms, flip angle 40°, 7 mm-cut edge and a 128 × 128 acquisition matrix (plane resolution 1.8 mm × 1.8 mm) with a 14 cm-caudal cerebellar coverage (20 cuts). The dynamic study consisted of a total 40 sequential volumes with an acquisition time for each one of 2.4 s. The administration of contrast was done through an infusion bomb using the antecubital vein as the IV – characterized as a 18 G cannula. Gadodiamide was used as the contrast agent (Omniscan®, GE Healthcare, U.S.A.) in a 0.2 mmol/kg dose and at an infusion speed of 5 ml/s. It was completed through a piston-driven pump of 30 ml physiological serum at the same flow. The administration of contrast was launched after initiating the acquisition of the third-row dynamic to allow the stabilization of the sequence signal.

The acquired images were transferred to a workstation to be further processed using a software designed and developed with Matlab® R2006b (MathWorks Inc., Natick, MA, U.S.A.).
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cerebral artery that was best seen in the measurement of the arterial input functions was chosen reducing the possibility of sample errors. ROI were defined manually coinciding with the tumor and peritumor areas in all consecutive cuts in which a lesion could be identified. Then all regions selected were linked to measure the volumes of interest. The necrotic components were included in the volumes demarcated by the analysis yet they did not contribute to the results since the perfusion values obtained are null and were not averaged with the rest.

In all enhanced lesions the tumor area was defined as the region with a signal enhancement in T1-weighted sequences after the administration of contrast. In the case of no relevantly enhanced lesions the tumor was defined as the region with signal alterations in the T2-weighted and FLAIR sequences. The white matter substance surrounding up to 1 cm of maximum distance in the region defined as tumor (Fig. 1) was considered the peritumoral area. Even though these regions do not rule out tumor infiltration or the inclusion of healthy tissue into the peritumoral region they are definitions that with certain variations are used in other works for the grading of tumors through MRI perfusion. In the cases of patients with mulcentric lesions the biopsied lesion was used for analysis only.

Two radiologists with at least 5-year experience in cerebral perfusion using MRI and not knowing the histopathological result of the tumor degree selected the ROI by consensus excluding those cerebral vessels that might alter the results of quantification. The selection of ROI was done directly on the perfusion images serving the morphological sequences of contrast-enhanced T1, T2 and FLAIR-weighted sequences (Fig. 1) orientative reference. The models used to quantify perfusion are now described briefly.

Monocompartmental model
The signal variations seen in the perfusion studies are due to effects of relaxativiy-based contrast agent and show the combination of the first step kinetic approach and its extravasation to the interstitial space. This model assumes that the recirculation of contrast and its extravasation are insignificant. However in brain tumors patency is usually enhanced which is a bias in the interpretation of the results. In our case to minimize this effect we have corrected the uptake curves by eliminating the recirculation and extravasation stages leaving the vascular stage only. BV, MTT, and BF parameters were measured too.

Pharmacokinetic or bicompartmental model
The pharmacokinetic model is based on the adjustment of the uptake curves to the contrast interchange between the intravascular compartment and the extracellular interstitial. The pharmacokinetic parameters used were the $K^{trans}$, $v_p$, $v_e$, and $k_{ep}$ coefficients. To obtain each and everyone of these parameters the artery and tissue response curves were adjusted to a mathematical model. Unlike the monocompartmental model no prior adjustments were made to correct the effect of recirculation since we are taking into account the first step, the following steps, and the contrast media clearance.

**Figure 1** Selection of the different regions of interest (ROI) from morphological and perfusion sequences. Middle cerebral artery, tumor and peritumoral regions. Morphological images served as orientative reference to later adjust the regions to perfusion images on which the analysis was carried out. (A and B) Enhanced tumoral lesion. Contrast material-enhanced T1-weighted sequence and perfusion image for a grade IV-glioma located in the right temporal lobe. (C–E) Non-enhanced tumor lesion. Contrast material-enhanced T1-weighted sequence, T2-weighted sequence and perfusion image for a grade II-glioma located in the left temporal lobe.

**Image analysis**

In all studies the middle cerebral artery (Fig. 1) was demarcated manually to extract the arterial input functions necessary to obtain perfusion parameters. The middle cerebral artery that was best seen in the measurement of the arterial input functions was chosen reducing the possibility of sample errors. ROI were defined manually coinciding with the tumor and peritumor areas in all consecutive cuts in which a lesion could be identified. Then all regions selected were linked to measure the volumes of interest. The necrotic components were included in the volumes demarcated by the analysis yet they did not contribute to the results since the perfusion values obtained are null and were not averaged with the rest.

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Statistical analysis

The ANOVA test was used to analyze the average, standard deviation (SD), and kurtosis of volume distribution corresponding to the tumor and peritumoral areas. SD and kurtosis were considered quantitative measurements of heterogeneity of the analyzed region. SD shows the dispersion of distribution values while kurtosis measures the shape of its histogram in such a way that a peaked histogram with a distribution of values very well centered around average shows a greater kurtosis than an histogram with dispersion of values.

To focus the analysis on the areas representing the greatest alteration in perfusion another ANOVA test was performed taking the maximum 10% of distributions. However in this case the measurement of kurtosis was ruled out given it could be a significant bias if not measured on the complete histogram of distribution.

Then we did post hoc analyses of each parameter for the evaluation of multiple comparatives through the Bonferroni method. We also did discrimination analyses to see if the linear combination of certain parameters improved the individual categorization. To build the classifiers of discrimination analyses the leave-one-out method was used. \( P < 0.05 \) value was considered statistically significant.

To evaluate the sensibility and specificity of all measurements the ROC curves were obtained by dividing patients into low-grade (II) and high-grade (III and IV) patients. The optimal cut-off values, that is, those showing the greatest sensibility and specificity were obtained from a graphical analysis of the curve in which each cut-off value (in the x axis), its sensibility and specificity (in the y axis) are represented being the cut-off value in which both curves meet the chosen one.

Results

At the tumor volume the statistically significant differences between the three tumor degrees for \( k^{\text{trans}} \), \( V_p \), \( V_e \), and \( k_{ep} \) and the corresponding SD were obtained both for the complete distribution and for the maximum 10% of histogram (Tables 1 and 2). The statistical significance of the results obtained was slightly higher for the complete distribution. For the 10% study statistically significant differences too were obtained for \( V_e \). The remaining parameters including the kurtosis values showed no statistically significant differences.

At the peritumoral area no statistically significant differences were obtained for any of the studied parameters in the study of the complete distribution of the histogram or in the maximum 10%. Only trends to the increase of individually tailored values could be observed.

In the study of heterogeneity a significant increase of SD of the individually tailored distributions could be seen with the increase of tumor degree. In Fig. 2 we can see this heterogeneity in the parametric mapping of BV. Contrary to what we could have expected kurtosis did not show up—not even a diminishing trend with tumor degree.

The multiparametric discriminating analysis improved the ability to categorize tumors with respect to classification rates of individual parameters. In particular the number of high-degree tumors erroneously classified as low-degree tumors was reduced yet we still had a 31% rate of error of grade III tumors categorized as grade II and a 5% rate of error of grade IV tumors categorized as grade II. Individually the average value of BV was the parameter that best categorized tumor grades showing a rate of error of 39% in grade III tumors categorized as grade II and a 13% rate of grade IV tumors categorized as grade II.

In the ROC curves areas >90% could be obtained for all statistically significant parameters (Fig. 3). From these curves the corresponding values of sensibility and specificity and the cut-off values for every variable (Table 3) too could
**Table 1** Results from the study of perfusion parameters, standard deviation and kurtosis comparing the degrees for the tumoral and peritumoral volumes using the complete histogram (average ± standard deviation).

<table>
<thead>
<tr>
<th></th>
<th>Tumoral</th>
<th>Peritumoral</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Grade II</td>
<td>Grade III</td>
</tr>
<tr>
<td>$k_{\text{trans}}$</td>
<td>0.01 ± 0.01</td>
<td>0.12 ± 0.13</td>
</tr>
<tr>
<td>$k_{\text{ep}}$</td>
<td>1.92 ± 4.14</td>
<td>5.08 ± 7.60</td>
</tr>
<tr>
<td>$v_e$</td>
<td>0.004 ± 0.001</td>
<td>0.004 ± 0.003</td>
</tr>
<tr>
<td>$v_p$</td>
<td>0.001 ± 0.001</td>
<td>0.002 ± 0.004</td>
</tr>
<tr>
<td>MTT</td>
<td>13.7 ± 2.6</td>
<td>21.0 ± 18.6</td>
</tr>
<tr>
<td>BV</td>
<td>0.01 ± 0.00</td>
<td>0.02 ± 0.02</td>
</tr>
<tr>
<td>BF</td>
<td>0.003 ± 0.001</td>
<td>0.006 ± 0.004</td>
</tr>
<tr>
<td>$D_{\text{trans}}$</td>
<td>0.03 ± 0.03</td>
<td>0.17 ± 0.14</td>
</tr>
<tr>
<td>$D_{\text{ep}}$</td>
<td>46.8 ± 140.6</td>
<td>6.5 ± 8.3</td>
</tr>
<tr>
<td>$D_e$</td>
<td>0.002 ± 0.001</td>
<td>0.005 ± 0.003</td>
</tr>
<tr>
<td>$D_v$</td>
<td>0.000 ± 0.001</td>
<td>0.001 ± 0.002</td>
</tr>
<tr>
<td>$D_{\text{MTT}}$</td>
<td>2.3 ± 1.9</td>
<td>6.9 ± 10.5</td>
</tr>
<tr>
<td>$D_{\text{BV}}$</td>
<td>0.004 ± 0.001</td>
<td>0.014 ± 0.011</td>
</tr>
<tr>
<td>$D_{\text{BF}}$</td>
<td>0.001 ± 0.000</td>
<td>0.004 ± 0.003</td>
</tr>
<tr>
<td>$C_{\text{trans}}$</td>
<td>21.5 ± 22.6</td>
<td>20.3 ± 28.8</td>
</tr>
<tr>
<td>$C_{\text{ep}}$</td>
<td>41.9 ± 40.0</td>
<td>15.7 ± 30.5</td>
</tr>
<tr>
<td>$C_v$</td>
<td>17.2 ± 22.0</td>
<td>16.1 ± 27.0</td>
</tr>
<tr>
<td>$C_p$</td>
<td>11.4 ± 20.3</td>
<td>20.8 ± 27.3</td>
</tr>
<tr>
<td>$C_{\text{MTT}}$</td>
<td>11.1 ± 18.1</td>
<td>13.5 ± 12.0</td>
</tr>
<tr>
<td>$C_{\text{BV}}$</td>
<td>10.6 ± 16.5</td>
<td>6.1 ± 2.9</td>
</tr>
<tr>
<td>$C_{\text{BF}}$</td>
<td>7.4 ± 8.2</td>
<td>9.0 ± 10.7</td>
</tr>
</tbody>
</table>

BF, blood flow; $k_{\text{ep}}$, cleansing coefficient; $k_{\text{trans}}$, transfer coefficient; MTT, mean transit time; $v_e$, interstitial volume; $v_p$, vascular volume; BV, blood volume. $D$ indicates the statistics of the standard deviation. Units: $k_{\text{trans}}, k_{\text{ep}},$ BF (s$^{-1}$), MTT (s), $v_e$, $v_p$, BV (score for one, no units).

$^*$ Significant differences between grades II and III. $^*$ Significant differences between grades II and IV.
Table 2  Results from the study of perfusion parameters, standard deviation and kurtosis comparing the degrees for the tumoral and peritumoral volumes using the maximum 10% (average ± standard deviation).

<table>
<thead>
<tr>
<th></th>
<th>Tumoral</th>
<th>Peritumoral</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Grade II</td>
<td>Grade III</td>
</tr>
<tr>
<td>$k_{\text{trans}}$</td>
<td>0.12 ± 0.21</td>
<td>0.47 ± 0.41</td>
</tr>
<tr>
<td>$k_{\text{ep}}$</td>
<td>16.2 ± 38.0</td>
<td>15.3 ± 19.0</td>
</tr>
<tr>
<td>$v_e$</td>
<td>0.008 ± 0.004</td>
<td>0.014 ± 0.007</td>
</tr>
<tr>
<td>$v_p$</td>
<td>0.001 ± 0.002</td>
<td>0.005 ± 0.009</td>
</tr>
<tr>
<td>MTT</td>
<td>18.4 ± 5.1</td>
<td>34.9 ± 37.3</td>
</tr>
<tr>
<td>$D_{\text{trans}}$</td>
<td>0.02 ± 0.02</td>
<td>0.05 ± 0.04</td>
</tr>
<tr>
<td>$D_{\text{ep}}$</td>
<td>0.006 ± 0.004</td>
<td>0.014 ± 0.009</td>
</tr>
<tr>
<td>$D_{\text{me}}$</td>
<td>0.05 ± 0.06</td>
<td>0.15 ± 0.11</td>
</tr>
<tr>
<td>$D_{\text{op}}$</td>
<td>3.0 ± 4.6</td>
<td>3.1 ± 6.1</td>
</tr>
<tr>
<td>$D_{\text{MTT}}$</td>
<td>0.002 ± 0.002</td>
<td>0.001 ± 0.010</td>
</tr>
<tr>
<td>$D_{\text{kurt}}$</td>
<td>0.000 ± 0.001</td>
<td>0.001 ± 0.002</td>
</tr>
<tr>
<td>$D_{\text{BV}}$</td>
<td>2.7 ± 4.6</td>
<td>4.8 ± 4.1</td>
</tr>
<tr>
<td>$D_{\text{BF}}$</td>
<td>0.00 ± 0.00</td>
<td>0.01 ± 0.01</td>
</tr>
<tr>
<td>$D_{\text{VF}}$</td>
<td>0.00 ± 0.001</td>
<td>0.003 ± 0.003</td>
</tr>
</tbody>
</table>

BF, blood flow; $k_{\text{ep}}$, cleansing coefficient; $k_{\text{trans}}$, transfer coefficient; MTT, mean transit time; $v_e$, interstitial volume; $v_p$, vascular volume; BV, blood volume. D indicates the statistics of the standard deviation. Units: $k_{\text{trans}}$, $k_{\text{ep}}$, BF (s$^{-1}$), MTT (s), $v_e$, $v_p$, BV (score for one, no units).
* Significant differences between grades II and III and grades II and IV.
** Significant differences between grades II and IV.
Table 3  Cut-off values and the corresponding sensibility and specificity values obtained from ROC curves of BV, BF, Ktrans and its standard deviations.

<table>
<thead>
<tr>
<th></th>
<th>Cut-off value</th>
<th>Sensibility (%)</th>
<th>Specificity (%)</th>
</tr>
</thead>
<tbody>
<tr>
<td>BV</td>
<td>0.013</td>
<td>86</td>
<td>90</td>
</tr>
<tr>
<td>BF</td>
<td>0.0035</td>
<td>88</td>
<td>90</td>
</tr>
<tr>
<td>Ktrans</td>
<td>0.027</td>
<td>82</td>
<td>80</td>
</tr>
<tr>
<td>D_BV</td>
<td>0.0065</td>
<td>90</td>
<td>90</td>
</tr>
<tr>
<td>D_BF</td>
<td>0.0018</td>
<td>90</td>
<td>100</td>
</tr>
<tr>
<td>D_Ktrans</td>
<td>0.08</td>
<td>82</td>
<td>80</td>
</tr>
</tbody>
</table>

BF, blood flow; Ktrans, transfer coefficient; BV, blood volume.

Units: BV (score for one, no units), BF, and Ktrans (s⁻¹). D indicates the statistics of the standard deviation.

Discussion

In our series the quantification of the average value of perfusion parameters of the monocompartmental module, BV, BF, the pharmacokinetic model, and the Ktrans coefficient—all obtained at the tumor volume—allowed us to statistically categorize degrees. These results correlate to other studies in which a good correlation between VScr and Ktrans values and tumor degree can be obtained. Unlike these and in an effort to increase the information collected from perfusion parameters we have used the quantification of tumor volume and histogram analyses vs the usual method of normalized ROI to the healthy white matter substance. However if we compare the ROC curves obtained with the ones published in those studies that have only used the relative value of the areas of interest for categorization, the analysis of the complete tumor volume does not have an effect in the substantial improvement of results for the categorization of tumors. Yet despite using all cut volume where the lesion is located this result is similar to that of Law et al.’s study though they use a single cut and share individual ROIs vs the complete histogram of tumor region of such a cut.

Significant differences were seen between grade II tumors and the other two grades. As it has happened in other studies this means that with a grouping of low- (II) and high-grade cases (III and IV) these differences would have been kept being more evident.

On the anatomopathological level astrocytomas are infiltrating tumors and this is why peritumoral regions show neoplastic invasion of different degree with vascular neoformation of endothelial structures of heterogeneous distribution and greater patency. These histological changes are represented in the perfusion studies and can be useful for categorization purposes. Contrary to what we could have expected the perfusion data obtained in the peritumoral regions did not allow us to distinguish lesions in a statistically significant way yet we saw that the higher the tumor degree is the higher date are as well. Few studies have evaluated the peritumoral area for the degree-categorization of astrocytomas showing different results too. Young et al. showed that the BV, BF histogram analysis of the peritumoral area is superior to the measurements obtained both in the tumoral and total areas including the tumoral and peritumoral areas. Now they define the peritumoral area in a semiautomatic way such as the expansion of tumoral area with a 6 pixel-radius in such a way that they are included in
the physiological vessel and gray matter areas which theoretically speaking would mean that the range of values increases. Same as it happens in our study other authors cannot find any differences with which categorize them using the peritumoral region. Alternatively the study of peritumoral area has proven to be useful to differentiate between high-degree glial tumors and metastatic lesions. 

In our series the perfusion parameters SD of the 2 models used allowed us to categorize tumor grades. The use of SD as descriptor of heterogeneity and tumor gradation is subject to limitations associated with the sample size. Now in high-degree tumors if the region selected for study has a small size the SD can be small as well whereas by using the whole tumor volume for the acquisition of data there would be a more significant deviation showing the dispersion of values. On the other hand this decision minimizes the importance of the highest data that are the ones that would be associated with vascular proliferation and high tumor degree. We have seen that SD, BV, BF and $K_{\text{trans}}$ allowed us to make distinctions on tumor degrees from the statistical point of view. As a matter of fact the BF deviation gives us the greatest rates of sensibility and specificity in such a way that we are able to separate between high- and low-degree tumors and even make comparisons among the average values of BV, BF and $K_{\text{trans}}$. The other statistical descriptor we used--kurtosis--did not allow us to categorize tumor degrees in the tumor region in any of the perfusion parameters.

Other studies also dealt with the heterogeneity of monocompartmental perfusion parameters while trying to establish the degree of astrocytomas. For example, Law et al. used the analysis of histograms of VSC maps in gliomas to determine how efficient was it in gradation. As it happens in our study SD correlated to tumor degree but not kurtosis. Even though in both studies the utility of SD and the kurtosis of the perfusion parameter histograms for gradation have been studied there are methodological differences including the perfusion model used, the definition of ROI, and the area or volume the data were obtained from. Lupo et al. studied the heterogeneity of perfusion parameters in different regions but only in high-grade gliomas (III and IV) by using both the maximum peak of histogram and the percentage of recovery through the monocompartmental model. In their study the spatial distribution of the average tumor microvascularization measured using MRI perfusion showed a significant heterogeneity within regions and for each and everyone of the 2 tumor degrees. Both in those articles and in the results from our studies measuring the heterogeneity of perfusion values of astrocytomas shows its histological polymorphism which in turn helps obtain its pre-surgical gradation and opens the possibility that it can be used in the follow-up of patients for the assessment of any modifications that might occur in heterogeneity after anti-angiogenic therapies.

In the study of the 10% of maximum values significant differences in the same parameters than in the overall histogram study occur. When it comes to relevance results do not change substantially but $v_e$ ends up showing significant differences. These results correlate with other studies in which the pharmacokinetic model of first step showed a significant increase of $v_e$ correlated with $K_{\text{trans}}$ as $K_{\text{trans}}$ gradually increases. This feature can explain why $v_e$ shows statistical significance when the analysis focuses on regions with greater BV and $K_{\text{trans}}$ values.

Usually the analysis of the patency of tumor vessels represented by $K_{\text{trans}}$ is done using the dynamic contrast material-enhanced T1-weighted modality (DCE-T1)–considered the reference modality. However several studies talk about the use of the dynamic-enhanced T2*-weighted modality. Our sequence allows us to adjust the pharmacokinetic calculations that are normally used with T1 enhancers given its quality of early resolution (2.4 s), total sequencing (90 s) and standardization of the relationship between signal and concentration. However it is worth noting that with a short acquisition time below 2 min, it is possible that the slowest stages of extravasation and cleansing are not adequately quantified.

Our study has several limitations. The number of cases included in the study is relatively small—similar to other studies focused on the gradation of astrocytomas though. The most represented group in our series is grade IV (64%) and the least represented of all grade II (16%). Nevertheless we believe that degree distribution allows us to be confident when analyzing the differences. Another limitation of our study is that of the method used for tumor grading such as the anatomopathological diagnosis. We know that astrocytomas can have different degrees in different regions of the tumor and this is why the sample used for the anatomopathological diagnosis cannot be fully representative. In our series we got to the diagnosis through the biopsy of 41% of the cases. This pattern of reference could have biased the results due to the partial sample of tumors vs the analysis of in vivo images of the total tumor volume perfusion. However it is likely that this bias will not affect results significantly in the whole study sample. On the other hand the perfusion parametric mapping obtained on complete volumes will allow us to focus the biopsies on those regions likely to show high degrees. Another aspect we should take into account is the input function. Some studies used a fixed standard function—even a vein some times as the vascular input function. As the individualized function we used the middle cerebral artery because it is a vessel identified in all studies. It is worth noting that in patients with localized tumors in the occipital lobe dependent on posterior circulation there might be a bias. Lastly the data have been obtained based on our definition of tumoral and peritumoral regions which might actually overlap in reality and bias the results. As a matter of fact the definition of peritumoral area is subject to controversy and varies from one study to the other. Regardless of how areas are defined the true limits of high-degree tumors are not confined in IV contrast-enhanced regions or in signal alteration areas but they spread to regions that look apparently healthy on images.

In sum the quantitative parameters obtained using the volume of data on the tumoral region through the monocompartmental model—the BV and the BF—and the volume of data obtained through the pharmacokinetic model--$K_{\text{trans}}$ are useful for the categorization of astrocytomas into different degrees. On the other hand the peritumoral region they are not useful yet they still show a certain trend to be high showing the alteration due to neoplastic infiltration. Heterogeneity—represented by standard deviation (SD) is also useful for categorization being the SD of the BF the most sensitive and specific parameter to distinguish...
between low- and high-grade tumors. The multiparametric discriminating analysis improved the ability to categorize tumors but not significantly. The information that perfusion studies using MRI give us for the grading of astrocytomas is useful but has limited value. However they are probably representing other aspects of tumor physiology valid to predict the prognosis and aggressiveness and are useful for the monitoring of tumor response to therapy.

**Ethical responsibilities**

**Protection of people and animals.** Authors confirm that the proceedings followed abide by the ethical regulations of the corresponding human experimentation committee in full compliance with the World Health Organization and the Declaration of Helsinki.

**Data confidentiality.** Authors confirm that in this report there are no personal data from patients.

**Right to privacy and informed consent.** Authors confirm that they have obtained the written informed prior consent from patients and/or subjects appearing in this article. This document is in the possession of the corresponding author.

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**Conflicts of interest**

Authors reported no conflicts of interest.

**References**


