Quantitative Assessment of Cerebral Blood Volume by Single-Photon Emission Computed Tomography

Umberto Sabatini, MD; Pierre Celsis, MD; Gérard Viallard, MSC; André Rascol, MD; and Jean-Pierre Marc-Vergnes, MD, PhD

We implemented a technique for measuring regional cerebral blood volume using single-photon emission computed tomography and in vivo technetium-99m-labeled red blood cells and then evaluated it in nine normal human volunteers (controls) and seven patients with bilateral occlusion or severe stenosis of the internal carotid artery. We also measured regional cerebral blood flow using single-photon emission computed tomography and intravenous xenon-133 in the same subjects. We studied regional cerebral blood flow, regional cerebral blood volume, and their ratio before and after the intravenous injection of 1 g acetazolamide. Mean±SD baseline regional cerebral blood volume was higher in the patients than in the controls (4.1 ±0.6 versus 3.2 ±0.3 ml/100 g, p < 0.01), and mean±SD baseline regional cerebral blood flow was lower in the patients than in the controls (40.5 ±11 versus 55.6 ±11 ml/100 g/min, p < 0.05). Acetazolamide induced similar mean±SD increases in regional cerebral blood volume in both the controls and the patients (0.3 ±0.1 and 0.3 ±0.2 ml/100 g), while the mean±SD regional cerebral blood flow reactivity was significantly less in the patients than in the controls (12.6 ±7.6 versus 24.5 ±9.6 ml/100 g/min, p < 0.05). Our study shows that single-photon emission computed tomography can provide quantitative estimates of both regional cerebral blood volume and regional cerebral blood flow in humans. (Stroke 1991;22:324-330)

Regional cerebral blood volume (rCBV), regional cerebral blood flow (rCBF), and regional oxygen extraction fraction (rOEF) have been used to study cerebral hemodynamics by using positron emission tomography (PET) in patients with carotid occlusive disease.1-3 These studies show that an increased rCBV and a decreased rCBF/rCBV ratio are correlated with an increased rOEF. Thus, rCBV and the rCBF/rCBV ratio could be considered indexes of the cerebral hemodynamic reserve, that is, the capacity of the cerebral vessels to dilate in response to a decrease in perfusion pressure.2,3 However, routine measurement of rCBV is limited by the expense and scarcity of PET facilities. As a result, the practicality of this measurement to assess the risk of stroke or the indications for surgery has not been investigated in long-term longitudinal studies of large groups of patients.4 Single-photon emission computed tomography (SPECT), which indirectly assesses the cerebral hemodynamic reserve by measuring the rCBF reactivity to acetazolamide,5,6 might prove more suitable than PET for such studies. Few quantitative rCBV measurements using SPECT have been reported to date,7-11 and the ability of SPECT to quantify rCBV accurately has been questioned.12,13 We assessed the reliability of SPECT to quantify rCBV, and we measured rCBV and rCBF before and after acetazolamide in normal volunteers and in patients with bilateral carotid artery disease.

Subjects and Methods

We studied nine healthy, right-handed volunteers (six men and three women, mean±SD age 55±8 years). All controls had normal clinical and biological examinations, computed tomograms (CT scan), and extracranial and intracranial Doppler examinations. We also studied seven male patients (mean±SD age 68±8 years) with bilateral occlusions or severe (>80%) stenoses of the internal carotid arteries (ICAs) (Table 1). All occlusions and stenoses of the ICAs were demonstrated with bilateral biplane conventional angiography. Five patients had no clinical history of stroke and a normal CT scan, and the other
two patients (6 and 7) had a clinical history of regressive left hemiparesis with a small right Sylvian hypodensity on CT scan.

The rCBV data were obtained as follows: a catheter was inserted in the antecubital vein of each arm of the subject. Stannous pyrophosphate was injected into the right catheter 45 minutes before data acquisition. Thirty minutes later, the subject’s red blood cells were labeled in vivo by an intravenous injection of 920 MBq technetium-99m pectechnetate ($^{99}$Tc). Immediately thereafter, the subject was placed in the tomo-graph (Tomomatic 64, Medimatic Inc., Copenhagen, Denmark) in the supine resting state with the eyes closed, and the head was positioned to obtain the same slices as for the xenon-133 ($^{133}$Xe) rCBF measurement (see below). Data acquisition started 15 minutes after the $^{99}$Tc injection. Data acquisition consisted of a sequence of eight images, each recorded over 3 minutes, so that the total duration was 24 minutes. One gram acetazolamide was injected at the end of the second image (sixth minute). Blood samples were collected from the catheter contralateral to the $^{99}$Tc and acetazolamide injections 2, 5, 11, 17, and 23 minutes after the beginning of data acquisition. These five blood samples were weighed and counted for 3 minutes in a well counter, and after correction for radioactive decay the activity was expressed as counts per minute (cpm) per milliliter. The mean activity of the three aliquots because accurate quantification of rCBV depends largely on the reliability of reconstructed values of the tracer’s concentrations. The first series consisted of using the tomograph to count the tracer concentrations in 15-cm-diameter cylindrical phan-toms filled with 2, 1.5, 1, and 0.5 μCi/ml solutions of $^{99}$Tc. Each measurement was repeated four times. The second series consisted of counting the tracer concentrations in a 15-cm-diameter phantom divided into two hemicylindrical chambers. The first chamber was filled with a 1 μCi/ml solution of $^{99}$Tc, and the other chamber was filled with solutions differing by 0%, 5%, 10%, and 15%. Each measurement was repeated three times. For both series, the $^{99}$Tc solutions in the phantom were chosen so as to give a total count in the reconstructed slice comparable to that obtained in the subjects (approximately 1.0 million cpm). In each case, weighed aliquots were counted in a well counter to obtain the actual concentrations.

The rCBF was measured the day before rCBV was assessed, using the same tomograph and intravenous injection of 2,200 MBq $^{133}$Xe. Data were obtained from three transverse slices 2 cm thick (in-plane resolution 1.7 cm), parallel and centered 1, 5, and 9 cm above the orbitomeatal plane. In this study, we considered only the middle slice because the first slice corresponds mainly to the cerebellar hemispheres, which do not pertain to the carotid territory.

TABLE 1. Left and Right rCBV and rCBF at Baseline and After Acetazolamide in Seven Patients With Bilateral Occlusions or Severe (>80%) Stenoses of Internal Carotid Arteries and Mean in Nine Controls

<table>
<thead>
<tr>
<th>Patients</th>
<th>rCBV Baseline (ml/100 g)</th>
<th>rCBF Baseline (ml/100 g/min)</th>
<th>rCBV After (ml/100 g)</th>
<th>rCBF After (ml/100 g/min)</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Left</td>
<td>Right</td>
<td>Left</td>
<td>Right</td>
</tr>
<tr>
<td>1</td>
<td>90</td>
<td>100</td>
<td>4.3</td>
<td>4.5</td>
</tr>
<tr>
<td>2</td>
<td>80</td>
<td>100</td>
<td>3.9</td>
<td>4.3</td>
</tr>
<tr>
<td>3</td>
<td>80</td>
<td>100</td>
<td>3.4</td>
<td>3.8</td>
</tr>
<tr>
<td>4</td>
<td>90</td>
<td>100</td>
<td>4.1</td>
<td>3.8</td>
</tr>
<tr>
<td>5</td>
<td>80</td>
<td>100</td>
<td>3.7</td>
<td>3.5</td>
</tr>
<tr>
<td>6*</td>
<td>80</td>
<td>100</td>
<td>5.2</td>
<td>5.2</td>
</tr>
<tr>
<td>7*</td>
<td>90</td>
<td>100</td>
<td>3.6</td>
<td>3.6</td>
</tr>
</tbody>
</table>

Controls (mean±SD) | 3.3±0.3 | 3.2±0.3 | 3.5±0.2 | 3.4±0.2 | 56±11 | 55±10 | 80.9±15 | 80.0±15 |

rCBV, regional cerebral blood volume; rCBF, regional cerebral blood flow.

*Symptomatic patients.

The rCBV in voxel i was calculated according to the classical formula as Ci×R/Cs×h, where Ci is the activity in voxel i (cpm), Cs is the activity concentration in the blood (cpm/ml), and h is the ratio of cerebral to large- vessel hematocrit, fixed at 0.85, the value currently used in the literature. The calibration factor, R, was obtained as follows: a 15-cm-diameter cylindrical phantom was filled with a 1 μCi/ml solution of $^{99}$Tc and counted for 3 minutes in the tomograph. The mean activity per voxel of the reconstructed image was determined. Three aliquots of the $^{99}$Tc solution were then withdrawn, weighed, and counted in the same well counter used to determine blood activity. The mean activity of the three aliquots was expressed as cpm per milliliter. After correction for radioactive decay, R was calculated as (mean count well counter) / (mean count voxel)×100/dt, where dt=1.05 g/ml (density of cere-bral tissue).

We performed two series of phantom measure-ments because accurate quantification of rCBV de-pends largely on the reliability of reconstructed values of the tracer’s concentrations. The first series consisted of using the tomograph to count the tracer concentrations in 15-cm-diameter cylindrical phan-toms filled with 2, 1.5, 1, and 0.5 μCi/ml solutions of $^{99}$Tc. Each measurement was repeated four times. The second series consisted of counting the tracer concentrations in a 15-cm-diameter phantom divided into two hemicylindrical chambers. The first chamber was filled with a 1 μCi/ml solution of $^{99}$Tc, and the other chamber was filled with solutions differing by 0%, 5%, 10%, and 15%. Each measurement was repeated three times. For both series, the $^{99}$Tc solutions in the phantom were chosen so as to give a total count in the reconstructed slice comparable to that obtained in the subjects (approximately 1.0 million cpm). In each case, weighed aliquots were counted in a well counter to obtain the actual concentrations.

The rCBF was measured the day before rCBV was assessed, using the same tomograph and intravenous injection of 2,200 MBq $^{133}$Xe. Data were obtained from three transverse slices 2 cm thick (in-plane resolution 1.7 cm), parallel and centered 1, 5, and 9 cm above the orbitomeatal plane. In this study, we considered only the middle slice because the first slice corresponds mainly to the cerebellar hemispheres, which do not pertain to the carotid territory.
FIGURE 1. Single-photon emission computed tomogram of regional cerebral blood volume in resting state from normal volunteer. Cortical/subcortical region of interest was defined in each hemisphere as shown on left side of image. Color scale is in milliliters/100 g.

and because data from the third slice are frequently contaminated by signal from the superior longitudinal sinus.

The subjects were positioned by means of an external removable grid. External landmarks were drawn on subject’s skin to facilitate repositioning for rCBV assessment. Two successive examinations were performed, one in the basal condition and the second 15 minutes after the injection of 1 g acetazolamide. The rCBF was quantified using an algorithm previously described. Because of the acetazolamide injection, rCBV could not be measured immediately after rCBF; rCBV was thus measured 24 hours later.

Image processing consisted of delineating the brain contour on the rCBV image and choosing identical regions of interest for the rCBV and rCBF images. For this purpose, we first superimposed the rCBF image on the rCBV image to delineate the rCBV brain contour and to exclude bone and the scalp. Afterwards, we delineated on both the rCBV and rCBF images a region of interest in each hemisphere so as to exclude contamination from the skull, scalp, dural sinuses, vessels of the sylvian fissure, ventricles (which in all cases were of normal size on a CT scan), and choroid plexuses (Figure 1). The region of interest was large enough to minimize intersubject variation in the gray/white matter ratio and to limit the importance of the partial volume effect. Hemispheric rCBV and rCBF values corresponded to the average of all voxels in the region of interest. Global mean rCBV and rCBF values corresponding to the average of both regions of interest were also calculated. The infarcted areas (defined on corresponding CT scans) in the two symptomatic patients were not excluded. Baseline rCBV was assessed using the data corresponding to the first image; rCBV after acetazolamide was measured using the data corresponding to the seventh image.

All values are expressed as mean±standard deviation (SD). Statistical significance was assessed using the nonparametric Wilcoxon and Mann-Whitney tests.

The study was approved by the Regional Ethics Committee. Informed consent was obtained from the subjects.

Results

The phantom study showed good reliability of the reconstructed concentrations compared with the actual concentrations and a good capacity of our technique to quantify small differences in the concentrations of two adjacent regions of a single reconstructed image (here the two chambers of the phantom, hence the two hemispheres of a subject as well) (Figure 2).

The 99mTc concentrations of the five blood samples collected during the 24 minutes of rCBV data acquisition remained stable; no significant difference was observed between activities of the first and last blood samples (7,201±2,835 versus 7,249±3,067 cpm). We observed no variation in PaCO2 during the 24 minutes of the rCBV examination, and there was no difference in PaCO2 between the controls and patients at baseline (35±4 versus 36±4 mm Hg). There was no difference between the beginning and end of the data acquisition sequence in mean ABP (controls: 95±9 versus 95±9 mm Hg; patients: 105±16 versus 101±14 mm Hg) or venous hematocrit (controls: 40±5% versus 39±4%; patients: 40±4% versus 42±2%). The patients’ mean ABP at rest was slightly higher than that of the controls, but this difference was not significant (105±16 versus 95±9 mm Hg). In the controls and the patients there was no significant difference in mean left and right rCBV and rCBF values before and after acetazolamide (Table 1). Therefore, the following results are expressed as the mean of both sides (Table 2).

Table 2 gives the mean values for rCBV, rCBF, and the rCBF/rCBV ratio in the controls and patients before and after acetazolamide injection. In the controls, acetazolamide significantly increased rCBV by 0.3 ml/100 g (9%, p<0.01) (Figure 3), rCBF by 24.8 ml/100 g/min (45%, p<0.01), and the rCBF/rCBV ratio by 5.7 min−1 (33%, p<0.01). In the patients, at baseline rCBV was higher than that in the controls (p<0.01), and rCBF and the rCBF/rCBV ratio were less than those in the controls (p<0.05 and p<0.01, respectively). Acetazolamide induced the same rCBV increase in the patients as it did in the controls (0.3 ml/100 g, 7%) (Figure 3). On the contrary, in the patients acetazolamide increased rCBF by only 12.6 ml/100 g/min (31%, p<0.05) and the rCBF/rCBV ratio by only 2.1 min−1 (20%,
FIGURE 2. Graphs of reliability (A) and sensitivity (B) of phantoms' reconstructed concentrations of 99mTc using tomographic system. In A, reconstructed concentrations were compared with concentrations measured with well counter (coefficients of variation of four sets of four measures were <0.5%, so corresponding points cannot be distinguished). In B, percentage differences in reconstructed concentrations were compared with percentage differences in concentrations measured with well counter (points are superimposed for second and third sets of measures).

FIGURE 3. Graph of mean left (○) and right (●) hemispheric cerebral blood volume (CBV) in nine normal volunteers (B) and in seven patients with bilateral internal carotid artery lesions (A) before and after intravenous injection of acetazolamide.

$p<0.05$). These increases were significantly smaller than those in the controls $(p<0.01)$.

No difference was observed in the values of rCBV, rCBF, and the rCBF/rCBV ratio of the two symptomatic patients compared with those in the five asymptomatic patients (Table 1).

When rCBF reactivity of the controls and the patients was plotted against baseline rCBV, we observed a significant correlation $(r=-0.53, p<0.05)$ (Figure 4); subjects with a high baseline rCBV had a small increase in rCBF after acetazolamide.

Discussion

Several methods for measuring rCBV have been proposed.7,19-22 However, apart from the PET tech-

<p>| TABLE 2. Mean rCBV, rCBF, and rCBF/rCBV Ratio in Patients and Controls at Baseline and After Acetazolamide |
|---------------------------------------------------|-----------------|-----------------|-----------------|</p>
<table>
<thead>
<tr>
<th></th>
<th>rCBV (ml/100 g)</th>
<th>rCBF (ml/100 g/min)</th>
<th>rCBF/rCBV ratio (min⁻¹)</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Baseline</td>
<td>After</td>
<td>Baseline</td>
</tr>
<tr>
<td>Patients</td>
<td>4.1±0.6*</td>
<td>4.4±0.6†</td>
<td>40.5±11‡</td>
</tr>
<tr>
<td>Controls</td>
<td>3.2±0.3</td>
<td>3.5±0.2§</td>
<td>55.6±11</td>
</tr>
</tbody>
</table>

rCBV, regional cerebral blood volume; rCBF, regional cerebral blood flow. Values are mean±SD.

*p<0.01, 0.05, respectively, different from control by Mann-Whitney test.

†‡p<0.05, 0.01, respectively, different from baseline by Wilcoxon's test.
nique, no method has been considered a reliable tool. Yet, Sakai et al., measured cerebral hematocrit, quantified rCBV using SPECT and 99mTc in normal healthy volunteers. The apparent reliability of these results led us to reappraise the use of SPECT for measuring rCBV in humans. Our data confirm that SPECT quantified rCBV, indicated rCBV reactivity to acetazolamide, and demonstrated increased rCBV values in patients with bilateral carotid artery disease.

In our controls, rCBV at rest was 3.2±0.3 ml/100 g. Previously published rCBV values obtained with PET and SPECT in normal human volunteers range from 3.2 to 5.1 ml/100 g. 1-3,10, 21-24 Our results are in this range but are low. Methodologic differences may account for this finding. Most published values of rCBV correspond to cortical regions of interest, while we considered a large, intrahemispheric region encompassing cortical as well as subcortical areas. The rCBV appears to be lower in subcortical than in cortical regions. Furthermore, in the study of Sakai et al. the region taken into account represented the mean of four horizontal slices (between 4.2 and 7.8 cm below the vertex) while we considered data from only one slice. Finally, we used the rCBF image as a mask to carefully delineate the cerebral tissue for rCBV measurement and to exclude any putative artifact due to bone or large vessels, which could increase the results. The second methodologic difference that could explain our low rCBV values is the age of our controls, which is greater than those in other works. Indeed, Leenders et al. reported that rCBV decreases with age.

The left hemispheric rCBV has been reported to be significantly greater than the right one in normal populations. This difference was said to correspond to the left–right asymmetry described in the human temporal cortex, which is thought to be greater in individuals with left cerebral dominance for speech. We did not observe the same difference, although mean rCBV of the left hemisphere was slightly larger than that of the right in our controls (Figure 3). This partly negative result can be explained by the fact that our regions of interest included cortical but also large subcortical areas. We observed, like Gibbs et al., that the mean baseline rCBV in patients with bilateral occlusions or severe stenoses of the ICAs was significantly higher than that in controls. This finding is in good agreement with the interpretative scheme proposed by Powers and Raichle.1 If the results of our patients are considered individually, four of them (1, 2, 4, and 6) exhibited an "abnormally" high rCBV at rest (i.e., higher than the control mean +2 SD). Interestingly, three of these patients (2, 4, and 6) showed the lowest rCBF values (Table 1). Conversely, the other three patients (3, 5, and 7) had moderately high rCBV values and moderately low rCBF values (within the normal range). This observation suggests that patients 2, 4, and 6 had already reached the limit of their cerebral hemodynamic reserve while patients 1, 3, 5, and 7 had not.

We also evaluated the rCBF/rCBV ratio, which is the reciprocal of the transit time and considered to be an index of the perfusion reserve. It has been suggested that this ratio is one of the most sensitive parameters for hemodynamic evaluation. In eight patients with bilateral ICA occlusions, using PET Gibbs et al. reported that the rCBF/rCBV ratio was significantly reduced. Using SPECT, we observed the same result in the same type of patients. In our controls the value of this ratio was higher than that in other studies; this difference can be explained by the methodologic reasons previously discussed for rCBV values and by the fact that rCBF in our subjects was higher than that in other studies.

After acetazolamide, mean rCBF and rCBV increased significantly in both controls and patients;
rCBV increased by 9% in our controls. This is in full agreement with the results of Sakai et al,8 who also observed an increase of approximately 10% after inhalation of 5% CO2. The effect of acetazolamide on rCBF has been proposed as a relevant method to evaluate the cerebral hemodynamic reserve.4,5 In our subjects, acetazolamide induced a smaller increase of rCBF in the patients. This suggests that the cerebral hemodynamic reserve was reduced in patients at an adaptive stage. Like others,1,11 we observed that the greater the baseline rCBV, the smaller the rCBF reactivity (Figure 4). Thus, direct measurements of rCBF at rest and rCBF reactivity after acetazolamide can be considered as basically equivalent in evaluating the cerebral hemodynamic reserve. Further studies are required to determine which method is more suitable for current practice.

Acetazolamide induced similar increases of rCBV in the controls and the patients. This finding suggests that the patients’ cerebral vessels, although already vasodilated at rest, retained a good capacity to increase their caliber, at least in response to an acute stimulation. This apparently normal capacity to vasodilate was, however, ineffective in maintaining a normal rCBF. Similar findings have been described in animal26,29 and theoretical30 models. In this situation, adaptive metabolic mechanisms (leading to an increase of rOEF) might already have been involved, as suggested by Kanno et al,31 and the adaptive mechanism schematized by Powers and Raichle1 should not be considered as fully sequential. Finally, the fact that the baseline rCBF/rCBV ratio was significantly smaller in our patients than in our controls suggests that, in patients with bilateral ICA occlusions or severe stenoses, the decreased perfusion pressure was the main factor responsible for the relative failure of the hemodynamic adaptive mechanism to maintain a normal rCBF. In conclusion, our results combined with those of other recent reports8–10 show that SPECT accurately quantifies both rCBF and rCBV and directly evaluates the cerebral hemodynamic reserve.

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References


KEY WORDS • cerebral blood flow • tomography, emission computed • acetazolamide • blood volume
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