

Original Article

Optimal timing for measuring cerebral blood flow after acetazolamide administration to detect preexisting cerebral hemodynamics and metabolism in patients with bilateral major cerebral artery steno-occlusive diseases: ^{15}O positron emission tomography studies

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Abstract: The present study determined the optimal timing of scanning for measurement of cerebral blood flow (CBF) after acetazolamide (ACZ) administration for detection of preexisting cerebral hemodynamics and metabolism in bilateral major cerebral artery steno-occlusive diseases. Thirty three patients underwent ^{15}O gas positron emission tomography (PET) and each parameter was obtained in the bilateral middle cerebral artery (MCA) territories. CBF was also obtained using H_2^{15}O PET scanning performed at baseline and at 5, 15, and 30 min after ACZ administration. Relative CBF at each time point after ACZ administration to baseline CBF was calculated. For MCA territories with normal cerebral blood volume (CBV) and cerebral metabolic rate of oxygen (CMRO_2), CBF continued increasing until 15 min after ACZ administration. For MCA territories with abnormally increased CBV, CBF decreased 5 min after ACZ administration. After that, CBF continued increasing until 30 min after ACZ administration. For MCA territories with abnormally decreased CMRO_2 , CBF did not change 5 min after ACZ administration. Ten min later, CBF increased. The accuracy to detect abnormally increased CBV was significantly greater for relative CBF_5 than for relative CBF_{15} . The accuracy to detect abnormally decreased CMRO_2 was significantly greater for relative CBF_5 or CBF_{15} than for relative CBF_{30} . For detecting abnormally increased oxygen extraction fraction, the accuracy did not differ among each relative CBF. These findings suggested that CBF measurement at 5 min after ACZ administration is the optimal timing for detection of preexisting cerebral hemodynamics and metabolism in bilateral major cerebral artery steno-occlusive diseases.

Keywords: Atherosclerotic steno-occlusive disease, acetazolamide, positron emission tomography, cerebral blood flow, steal phenomenon

Introduction

Stage 1 ischemia is the result of cerebrovascular autoregulatory mechanisms acting to preserve cerebral blood flow (CBF) through the dilation of precapillary resistance vessels following a reduction in cerebral perfusion pressure due to the chronic progression of atherosclerotic stenosis or occlusion in either the internal carotid artery (ICA) or middle cerebral artery (MCA), leading to an increase in cerebral blood volume (CBV) [1-3]. In stage 1 ischemia, cerebrovascular reactivity (CVR) to acetazol-

amide (ACZ), which indicates the degree of the cerebrovascular autoregulatory vasodilation, begins to decrease [1-3]. In stage 2 ischemia, autoregulatory capacity is unable to compensate for a further reduction in cerebral perfusion pressure, thereby resulting in reduced CBF and increased oxygen extraction fraction (OEF), also known as misery perfusion [1-3]. During this stage, CVR to ACZ disappears [1-3]. A further reduction in cerebral perfusion pressure finally leads to irreversible ischemic damage of neuronal tissue, resulting in a reduction in cerebral metabolism.

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Stage 2 ischemia, which is indicated by an increase in OEF on positron emission tomography (PET), increases the risk of stroke recurrence among patients with symptomatic arteriosclerotic occlusion of the ICA or MCA [4-6]. The incidence of recurrent ischemic events is low in adult patients with ischemic moyamoya disease without increased OEF who are receiving medication alone, and in such patients, increased OEF during the follow-up period is a strong predictor of recurrent ischemic events [7]. Therefore, identifying patients with increased OEF resulting from chronic steno-occlusive disease of the ICA or MCA could help to prevent stroke recurrence. In addition, some studies have reported that a preoperative increase in CBV in the cerebral hemisphere ipsilateral to carotid endarterectomy or the placement of a carotid artery stent for cervical ICA stenosis or arterial bypass surgery for moyamoya disease is an independent and powerful predictor of postoperative cerebral hyperperfusion [8-10], which can lead to the development of cerebral edema or intracerebral hemorrhage [8-10]. Even when asymptomatic, this phenomenon can cause slight but diffuse brain damage, thereby resulting in postoperative cognitive decline [11, 12]. Therefore, identifying increased CBV in such patients could provide improved risk stratification.

A study using ^{15}O PET for healthy adults demonstrated that CBF continued to increase until 10 min after and remained unchanged or decreased until 20 min after ACZ administration [13]. Another study using ^{15}O PET for patients with chronic progression of ICA or MCA steno-occlusive disease reported that CBF sometimes paradoxically decreased at 5 min after ACZ administration when compared with pre-administration, and at 15 min later, this decreased CBF increased over the pre-administration value [14]. Further, Okazawa et al. [15] used dynamic H_2^{15}O PET scans after ACZ injection in patients with unilateral major cerebral arterial occlusive disease and demonstrated that whereas CBF increased after ACZ administration in the cerebral hemisphere contralateral to the affected side, the ACZ administration sometimes caused no change or only a slight decrease in CBF in the affected cerebral hemisphere. These data suggest dynamic changes in CBF after ACZ administration and differences in such changes among patients and healthy

subjects. These data also suggest optimal timing in terms of scanning for the measurement of CBF after ACZ administration for the accurate detection of cerebral hemodynamics in patients with ICA or MCA steno-occlusive disease. In the clinical setting, while CVR to ACZ is measured using perfusion single-photon emission computed tomography (CT) [16], Xenon-enhanced CT [17], perfusion CT [18], or dynamic susceptibility contrast perfusion magnetic resonance imaging (MRI) [19], the timing of scanning for the measurement of CBF after ACZ administration varies among these modalities. In addition, cerebral hemodynamics are impaired more strongly or frequently in patients with bilateral atherosclerotic ICA or MCA steno-occlusive diseases compared with those with unilateral disease [20], and the optimal timing in terms of scanning for the measurement of CBF after ACZ administration in such patients also remains unknown.

Given this background, the present study aimed to determine the optimal timing of scanning for the measurement of CBF after ACZ administration for the detection of cerebral hemodynamics and metabolism in patients with bilateral atherosclerotic ICA or MCA steno-occlusive diseases by comparing dynamic changes in CBF after ACZ administration with preexisting cerebral hemodynamics and metabolism using ^{15}O PET.

Material and methods

All procedures in this study were carried out in accordance with the ethical standards of the ethics committee of our institute and the 1964 Declaration of Helsinki and its later amendments or comparable ethical standards. Institutional review board approval was obtained. Written informed consent was obtained from all participants or their next of kin before the study began.

Healthy subjects

We prospectively enrolled 10 healthy subjects who met the following inclusion criteria for obtaining normal control values of brain ^{15}O gas PET: age 30-70 years, no history of hypertension, diabetes mellitus, or dyslipidemia, and absence of leukoaraiosis or asymptomatic lacunar infarction on conventional brain MRI.

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Patients

We prospectively enrolled patients who met the following inclusion criteria: age 30-70 years, presence of clinical symptoms suggesting ischemic episodes in the MCA or ICA territory at 1-6 months prior to visiting our institute, useful residual function (score of 0, 1, or 2 on the modified Rankin scale), presence of bilateral atherosclerotic stenosis of intracranial ICA or the M1 of the MCA (greater than 70% or 50% for the ICA and MCA, respectively) or occlusion on cerebral angiography using arterial catheterization, CT, or MRI, and absence of infarcts in the entire cortical area supplied by the M4 branch of the MCA on T1-, T2-, and diffusion-weighted MRI.

Brain ^{15}O PET study

PET studies were performed using a 4-ring, 7-slice PET scanner (Headtome-IV; Shimadzu, Kyoto, Japan) with in-plane and axial resolutions of 8 mm and 10 mm, respectively [21]. This PET scanner provides 14 tomographic images with 6.5-mm intervals by the continuous axial motion of the gantry. The image slices were parallel to the orbitomeatal line. PET studies were performed in all patients more than 4 weeks after the last ischemic event. First, measurements of the cerebral metabolic rate of oxygen (CMRO_2) and OEF were obtained during continuous inhalation of $^{15}\text{O}_2$. Data were collected for 5 min. A single breath of C^{15}O was used to measure CBV. CMRO_2 and OEF were calculated using the steady-state method [22] and corrected by CBV [23]. Next, CBF was calculated using the autoradiography method with 90 s of scanning after an intravenous bolus injection of H_2^{15}O .²³ H_2^{15}O PET scanning was performed at the resting state (defined as baseline). ACZ (1000 mg, range 13-19 mg/kg body weight) was dissolved in physiological saline (20 mL), and 5 min after the end of baseline H_2^{15}O PET scanning, this solution was administered intravenously over a period of 1 min. The H_2^{15}O PET scanning was performed in the same fashion three times so that each mid-scan time was 5, 15, and 30 min after the end of intravenous administration of ACZ. Continuous arterial blood sampling and β -ray monitoring with a scintillator were conducted throughout PET scanning using a catheter implanted in the radial artery to obtain the arterial input function. Blood pH and arterial partial

pressure of both oxygen (PaO_2) and carbon dioxide (PaCO_2) were also measured in the same blood samples. Blood pressure was measured by auscultation during each CBF measurement. Healthy subjects and patients underwent PET studies with $^{15}\text{O}_2$ and C^{15}O in the same fashion.

Image data analysis

To ensure that the brain images of all participants would have the same anatomic format, the PET images were transformed into the standard brain size and shape by linear and nonlinear transformation for anatomic standardization using SPM2 software (Fujifilm RI Pharma, Tokyo, Japan) [25]. Next, using SPM2, a three-dimensional (3D) stereotaxic region of interest (ROI) template was used to place 318 constant ROIs in the cerebral and cerebellar hemispheres automatically [26]. Then, the ROIs were grouped into the following 10 segments in each hemisphere according to the arterial supply: callosomarginal, pericallosal, precentral, central, parietal, angular, temporal, posterior, hippocampal, and cerebellar. Next, five of the 10 segments-precentral, central, parietal, angular, and temporal-perfused by the MCA were combined, and this was defined as an MCA ROI. Mean values of CBV, OEF, CMRO_2 , and CBF were measured in the MCA ROIs in the bilateral cerebral hemispheres. In each MCA ROI of each cerebral hemisphere with ACZ challenge, relative CBF (%) was calculated as follows: $100 \times \text{CBF at each time point after ACZ administration} / \text{baseline CBF}$ (defined as relative CBF_5 , CBF_{15} , or CBF_{30}). Each MCA ROI of each patient was defined as having abnormally increased CBV or OEF when CBV or OEF was greater than the upper limit of the 95% confidence intervals (CIs) of the values obtained from 20 cerebral hemispheres of 10 healthy subjects, respectively. Conversely, each MCA ROI of each patient was defined as having abnormally decreased CMRO_2 when CMRO_2 was lower than the lower limit of the 95% CIs of values obtained from 20 cerebral hemispheres of 10 healthy subjects.

Statistical analysis

Data are expressed as the mean \pm standard deviation (SD) and/or the 95% CI. A chronological change in values at each time point during ACZ challenge was evaluated using repeated

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Table 1. Physiologic data during H₂¹⁵O PET scanning with acetazolamide administration in patients

	Baseline	5 min after ACZ administration	15 min after ACZ administration	30 min after ACZ administration
PaO ₂ (mm Hg)				
Mean	95.2	95.9	95.6	94.8
SD	3.1	3.8	4.1	4.3
PaCO ₂ (mm Hg)				
Mean	39.9	40.8	39.6	40.7
SD	3.0	2.8	3.1	2.7
pH				
Mean	7.39	7.40	7.41	7.40
SD	0.04	0.03	0.04	0.04
Mean blood pressure (mm Hg)				
Mean	97.6	98.5	96.8	97.0
SD	9.9	8.5	8.0	7.4

PET, positron emission tomography; ACZ, acetazolamide; SD, standard deviation.

measures analysis of variance. When this change was significant, differences among values at each time point were compared using Scheffé's test. A difference in values in three parameters or more was evaluated using multi-factor analysis of variance. When this difference was significant, differences among values in each parameter were compared using Scheffé's test. A difference in values in two parameters was evaluated using the Mann-Whitney *U* test. Statistical significance for all analyses was set at the *P*<0.05 level. The cutoff lying closest to the upper left corner of the receiver operating characteristic (ROC) curve for detecting abnormally increased CBV, abnormally increased OEF, or abnormally decreased CMRO₂ was determined for relative CBF at each time point after ACZ administration. Binomial distributions were used to calculate the exact 95% CIs for accuracy, sensitivity, specificity, and positive and negative predictive values, and differences in these values among relative CBF at each time point after ACZ administration were assessed using 95% CIs.

Results

Patient inclusion

Over the 36-month study period, a total of 36 patients who met the inclusion criteria were scheduled to undergo PET scanning. In two patients, PET scanning was canceled halfway through because of the development of claustrophobia. All of the remaining 34 patients successfully underwent PET scanning. However, in

one of these patients, data sufficient to generate imaging were not obtained because of motion artifacts; this patient was also excluded from the analysis. Therefore, 33 patients were finally enrolled in the study.

Clinical characteristics

Among these 33 patients (26 men, 7 women; mean age, 62±9 years; range, 37-69 years), 30 had hypertension, 10 diabetes mellitus, and 25 dyslipidemia. Fourteen and 12 patients had bilateral ICA and bilateral MCA steno-occlusive disease, respectively. The remaining seven patients had unilateral ICA and contralateral MCA steno-occlusive diseases. Therefore, 66 MCA ROIs of the 66 cerebral hemispheres were eventually analyzed. **Table 1** shows the physiologic data of the patients during H₂¹⁵O PET scanning with ACZ administration in the patients. No differences were seen in PaO₂, PaCO₂, blood pH, or mean blood pressure at baseline, 5, 15, or 30 min after ACZ administration.

Classification of ROIs based on preexisting cerebral hemodynamics

The mean age of the 10 healthy subjects (7 men, 3 women) who were enrolled to obtain normal brain ¹⁵O-gas PET values was 56±10 years (range, 35-68 years). **Table 2** shows the means and 95% CIs for CBF, CBV, OEF, and CMRO₂ in bilateral MCA ROIs in patients and healthy subjects. Based on data obtained from the healthy subjects, among 66 MCA ROIs in

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Table 2. Comparisons of CBV, OEF and CMRO₂ among MCA ROIs in patients and healthy subjects

	Patients (N=66)				A vs. B	A vs. C	A vs. D	B vs. C	B vs. D	C vs. D
	Healthy subjects (N=20)	Normal CBV and CMRO ₂ (N=41)	Abnormally increased CBV (N=15)	Abnormally decreased CMRO ₂ (N=10)						
	A	B	C	D						
CBF (mL/100 g/min)					0.0034	<0.0001	<0.0001	0.0076	0.0001	NS
Mean	44.9	39.4	34.2	32.9						
95% CI	35.5-54.3	30.2-48.5	27.8-40.5	23.5-42.1						
CBV (mL/100 g)					0.0181	<0.0001	0.0023	<0.0001	<0.0001	<0.0001
Mean	3.96	4.36	5.20	3.27						
95% CI	2.91-5.01	3.28-5.44	4.87-5.53	2.77-3.77						
OEF (%)					NS	<0.0001	NS	<0.0001	NS	0.0003
Mean	44.0	45.0	51.1	44.9						
95% CI	37.0-51.1	39.0-51.1	45.6-56.6	35.2-54.5						
CMRO ₂ (mL/100 g/min)					NS	NS	<0.0001	NS	<0.0001	<0.0001
Mean	3.28	3.12	3.25	2.22						
95% CI	2.27-4.29	2.28-3.96	2.81-3.69	2.13-2.31						

CBF, cerebral blood flow; CBV, cerebral blood volume; OEF, oxygen extraction fraction; CMRO₂, cerebral metabolic rate of oxygen; MCA, middle cerebral artery; ROI, region of interest; CI, confidence intervals; NS, not significant.

patients, 15 (23%) had abnormally increased CBV. The other 10 (15%) MCA ROIs had abnormally decreased CMRO₂. These 10 MCA ROIs did not exhibit abnormally increased CBV. The remaining 41 (62%) MCA ROIs had neither abnormally increased CBV nor abnormally decreased CMRO₂ (defined as normal CBV and CMRO₂). Eleven (17%) MCA ROIs had abnormally increased OEF: 9 and 2 exhibited abnormally increased CBV and abnormally decreased CMRO₂, respectively. CBF significantly differed among each ROI subgroup of patients and healthy subjects except the relationship between abnormally increased CBV and abnormally decreased CMRO₂, in the following descending order: healthy subjects > normal CBV and CMRO₂ > abnormally increased CBV = abnormally decreased CMRO₂. CBV significantly differed among each ROI subgroup of patients and healthy subjects in the following descending order: abnormally increased CBV > normal CBV and CMRO₂ > healthy subjects > abnormally decreased CMRO₂. OEF was significantly greater in an ROI subgroup with abnormally increased CBV than in the other two subgroups of patients and healthy subjects. OEF did not differ among any other combinations of ROI subgroups of patients and healthy subjects. CMRO₂ was significantly lower in an ROI group with abnormally decreased CMRO₂ than in the other two subgroups of patients and healthy subjects. CMRO₂ did not differ among any other combinations of ROI subgroups of patients and healthy subjects.

Chronological changes in CBF after ACZ administration

Figure 1 shows chronological changes in relative CBF after ACZ administration in each ROI subgroup with normal CBV and CMRO₂, abnormally increased CBV, and abnormally decreased CMRO₂ in patients. The CBF significantly changed in all ROI subgroups of patients (P < 0.0001). For an ROI subgroup with normal CBV and CMRO₂, CBF continued increasing until 15 min after ACZ administration (133.1±14.5% for relative CBF₅; 140.6±12.2% for relative CBF₁₅) and reduced 15 min later (126.8±13.5% for relative CBF₃₀). However, CBF₃₀ remained greater than baseline CBF. For the ROI subgroup with abnormally increased CBV, CBF decreased 5 min after ACZ administration (85.1±8.5% for relative CBF₅). After that, CBF continued increasing until 30 min after ACZ administration (104.3±12.7% for relative CBF₁₅; 110.0±10.3% for relative CBF₃₀). For the ROI subgroup with abnormally decreased CMRO₂, CBF did not change 5 min after ACZ administration (106.5±6.8% for relative CBF₅). Then, 10 min later, CBF increased (116.6±5.9% for relative CBF₁₅) and maintained this value until 30 min after ACZ administration (113.3±9.7% for relative CBF₃₀).

Comparisons of relative CBF at each time point after ACZ administration

Figure 2 shows comparisons of relative CBF at each time point after ACZ administration among each ROI subgroup with normal CBV and

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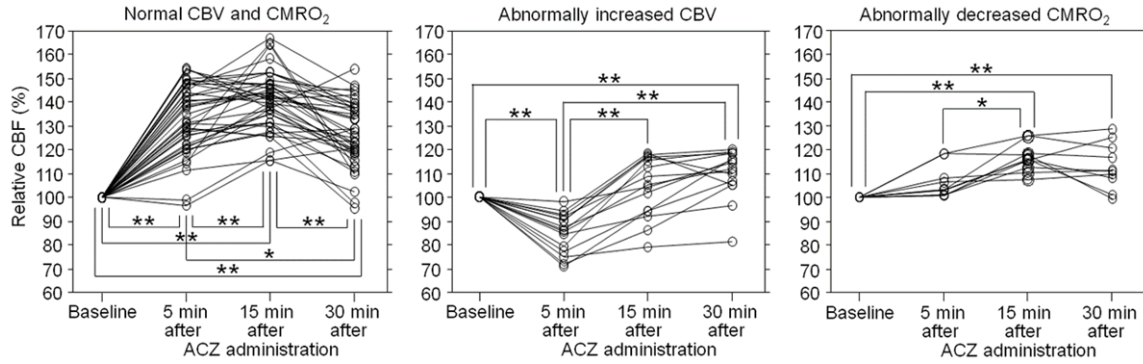


Figure 1. Chronological changes in relative cerebral blood flow (CBF) after acetazolamide (ACZ) administration in each region of interest (ROI) subgroup with normal cerebral blood volume (CBV) and cerebral metabolic rate of oxygen ($CMRO_2$), abnormally increased CBV, and abnormally decreased $CMRO_2$ in patients. * and ** denote $P<0.05$ and $P<0.01$, respectively.

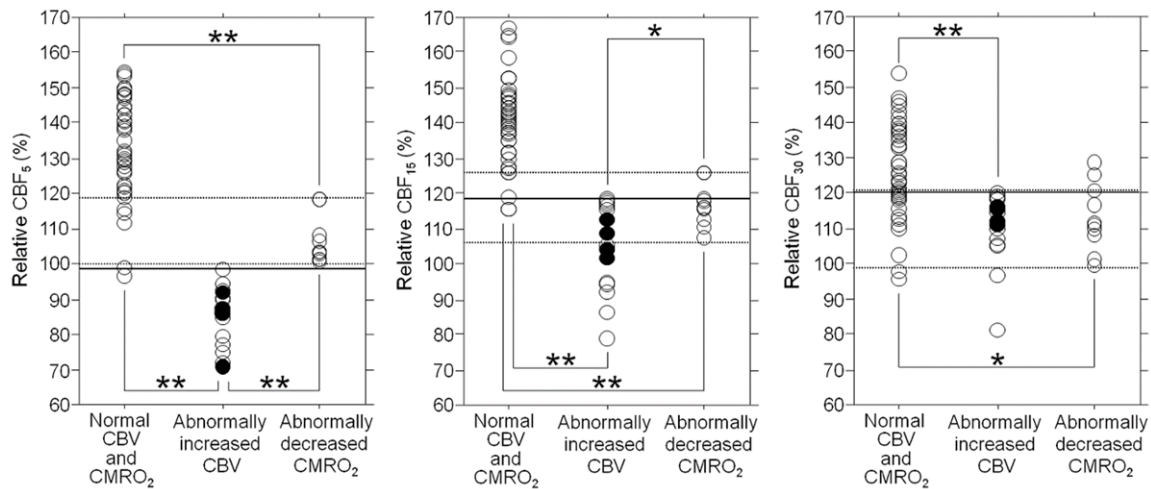


Figure 2. Comparisons of relative cerebral blood flow (CBF) at 5 (CBF_5), 15 (CBF_{15}), and 30 (CBF_{30}) min after acetazolamide (ACZ) administration among each region of interest (ROI) subgroup with normal cerebral blood volume (CBV) and cerebral metabolic rate of oxygen ($CMRO_2$), abnormally increased CBV, and abnormally decreased $CMRO_2$ in patients. Closed and open circles denote ROIs with and without abnormally increased oxygen extraction fraction (OEF), respectively. * and ** denote $P<0.05$ and $P<0.01$, respectively.

$CMRO_2$, abnormally increased CBV, and abnormally decreased $CMRO_2$ in the patients. Relative CBF_5 and CBF_{15} significantly differed among each ROI subgroup of patients and were in the following descending order: normal CBV and $CMRO_2 >$ abnormally decreased $CMRO_2 >$ abnormally increased CBV. Relative CBF_5 in all MCA ROIs with abnormally increased CBV exhibited $<100\%$. Relative CBF_{30} was significantly greater in the ROI subgroup with normal CBV and $CMRO_2$ than in the groups with abnormally increased CBV and abnormally decreased $CMRO_2$ and did not differ between the latter two ROI subgroups of patients. In 25 MCA ROIs with abnormally increased CBV or abnormally decreased $CMRO_2$, relative CBF at any time

point after ACZ administration did not differ between MCA ROIs with abnormally increased OEF (CBF_5 , $88.5 \pm 12.4\%$; CBF_{15} , $103.4 \pm 13.5\%$; CBF_{30} , $107.1 \pm 10.7\%$) and without abnormally increased OEF (CBF_5 , $97.6 \pm 12.8\%$; CBF_{15} , $112.7 \pm 8.7\%$; CBF_{30} , $114.5 \pm 8.3\%$).

Accuracy for relative CBF at each time point after ACZ administration for detecting preexisting cerebral hemodynamics

Table 3 shows the cutoff lying closest to the upper left corner of the ROC curve, the accuracy, sensitivity, specificity, and positive and negative predictive values for relative CBF at each time point after ACZ administration for detect-

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Table 3. Cutoff lying closest to the upper left corner of the ROC curve, accuracy, sensitivity, specificity, and positive- and negative-predictive values for relative CBF at each time point after ACZ administration for detecting abnormally increased CBV, abnormally increased OEF or abnormally decreased CMRO₂ in patients

		A	B	C	Statistical significance by comparison of 95% CI		
		Relative CBF ₅	Relative CBF ₁₅	Relative CBF ₃₀	A versus B	B versus C	C versus A
Abnormally increased CBV	Cutoff	98.6%	118.8%	120.0%			
	Accuracy	98%	85%	71%	Yes	No	Yes
	95% CI	96%-100%	76%-93%	60%-82%			
	Sensitivity	100%	100%	100%	No	No	No
	95% CI	100%-100%	100%-100%	100%-100%			
	Specificity	98%	80%	63%	Yes	No	Yes
	95% CI	93%-100%	69%-92%	49%-76%			
	Positive predictive value	94%	60%	44%	Yes	No	Yes
	95% CI	82%-100%	41%-79%	27%-61%			
	Negative predictive value	100%	100%	100%	No	No	No
95% CI	100%-100%	100%-100%	100%-100%				
Abnormally increased OEF	Cutoff	98.6%	115.6%	109.8%			
	Accuracy	89%	86%	85%	No	No	No
	95% CI	82%-97%	78%-95%	76%-93%			
	Sensitivity	91%	82%	64%	No	No	No
	95% CI	74%-100%	59%-100%	35%-92%			
	Specificity	89%	87%	89%	No	No	No
	95% CI	81%-97%	78%-96%	81%-97%			
	Positive predictive value	63%	56%	54%	No	No	No
	95% CI	39%-86%	32%-81%	27%-81%			
	Negative predictive value	98%	96%	92%	No	No	No
95% CI	94%-100%	91%-100%	85%-100%				
Abnormally decreased CMRO ₂	Cutoff	99.7 and 118.8	106.3 and 126.0	98.7 and 120.9			
	Accuracy	95%	83%	58%	No	Yes	Yes
	95% CI	90%-100%	74%-92%	46%-69%			
	Sensitivity	100%	100%	80%	No	No	No
	95% CI	100%-100%	100%-100%	55%-100%			
	Specificity	95%	80%	54%	Yes	Yes	Yes
	95% CI	91%-100%	70%-89%	41%-67%			
	Positive predictive value	77%	48%	24%	No	No	Yes
	95% CI	54%-100%	26%-69%	9%-38%			
	Negative predictive value	100%	100%	94%	No	No	No
95% CI	100%-100%	100%-100%	85%-100%				

ing abnormally increased CBV, and abnormally increased OEF or abnormally decreased CMRO₂ in the patients. Based on **Figure 2**, when the upper cutoff for detecting abnormally decreased CMRO₂ was determined, relative CBF in the ROI subgroup alone with normal CBV and CMRO₂ was used as a control, and when the lower cutoff for detecting abnormally decreased CMRO₂ was determined, relative CBF in the ROI subgroup alone with abnormally increased CBV was used as a control. For detecting abnormally increased CBV, the sensitivity and negative predictive value were 100% for

relative CBF₅, CBF₁₅, and CBF₃₀ and no differences were found among them; the accuracy, specificity, and positive predictive value were significantly greater for relative CBF₅ than for relative CBF₁₅ or CBF₃₀. For detecting abnormally increased OEF, no values differed among relative CBF₅, CBF₁₅, or CBF₃₀. For detecting abnormally decreased CMRO₂, the accuracy was significantly greater for relative CBF₅ and CBF₁₅ than for relative CBF₃₀; the sensitivity and negative predictive value did not differ among relative CBF₅, CBF₁₅, or CBF₃₀, the specificity significantly differed among relative CBF₅,

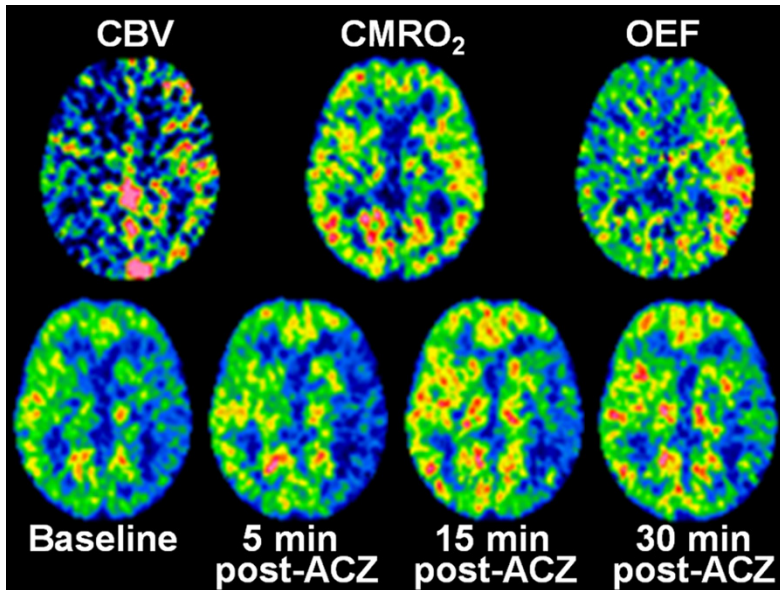


Figure 3. A 68-year-old man with transient ischemic attacks of right motor weakness due to left middle cerebral artery (MCA) occlusion and asymptomatic right MCA occlusion. In the symptomatic left MCA territory, baseline cerebral blood flow (CBF) was reduced, cerebral blood volume (CBV) and oxygen extraction fraction (OEF) were increased, and cerebral metabolic rate of oxygen (CMRO₂) was normal. In that territory, CBF was reduced at 5 min after ACZ administration when compared with baseline. Thereafter, CBF continued to increase until 30 min after acetazolamide (ACZ) administration. By contrast, in the asymptomatic right MCA territory, CBF continued to increase until 15 min after ACZ administration, and then decreased 15 min later.

CBF₁₅, and CBF₃₀ (CBF₅ > CBF₁₅ > CBF₃₀), and the positive predictive value was significantly greater for relative CBF₅ than for relative CBF₃₀.

Representative CBF images at baseline, 5, 15, and 30 min after ACZ administration and CBV, OEF, and CMRO₂ images in each patient with abnormally increased CBV (plus abnormally increased OEF) or with abnormally decreased CMRO₂ in the symptomatic cerebral hemisphere are shown in **Figures 3** and **4**, respectively.

Discussion

The present study demonstrated that CBF measurement at 5 min after ACZ administration is the optimal timing for detecting preexisting cerebral hemodynamics and metabolism in patients with bilateral atherosclerotic ICA or MCA steno-occlusive diseases.

Based on cerebral hemodynamics and metabolism, MCA territories in our patients were classified into abnormally increased CBV, abnormally decreased CMRO₂, and normal CBV and CMRO₂. None of MCA territories in the patients

exhibited both abnormally increased CBV and abnormally decreased CMRO₂. According to cerebrovascular autoregulatory mechanisms, precapillary resistance vessels dilate in response to a reduction in cerebral perfusion pressure, resulting in increased CBV [1-3]. This response may be due to chemical reactions [1-3] that depend on cerebral metabolism. Therefore, dilation of precapillary resistance vessels may be difficult in neural tissues with reduced cerebral metabolism. By contrast, OEF was increased in MCA territories with abnormally decreased CMRO₂ or abnormally increased CBV. These findings were comparable with those previously reported [27]. OEF is theoretically a function of CMRO₂/CBF, and neural tissues with further reduced CBF compared with

reduced CMRO₂ might exhibit abnormally increased OEF without abnormally increased CBV.

Chronological changes in CBF after ACZ administration differed among three categories classified based on cerebral hemodynamics and metabolism. For MCA territories with normal CBV and CMRO₂, CBF continued increasing until 15 min after ACZ administration and decreased thereafter. For MCA territories with abnormally increased CBV, CBF transiently decreased at 5 min after ACZ administration and then CBF continued increasing until 30 min after ACZ administration. Chronological changes in CBF after ACZ administration in MCA territories with normal CBV and CMRO₂ corresponded with those in healthy subjects previously reported [13]. By contrast, the changes in MCA territories with abnormally increased CBV have sometimes been reported to occur in the cerebral hemisphere with chronic progression of ICA or MCA steno-occlusive disease [14, 15]. In the present study, CBF was lower in MCA territories with abnormally increased CBV than in

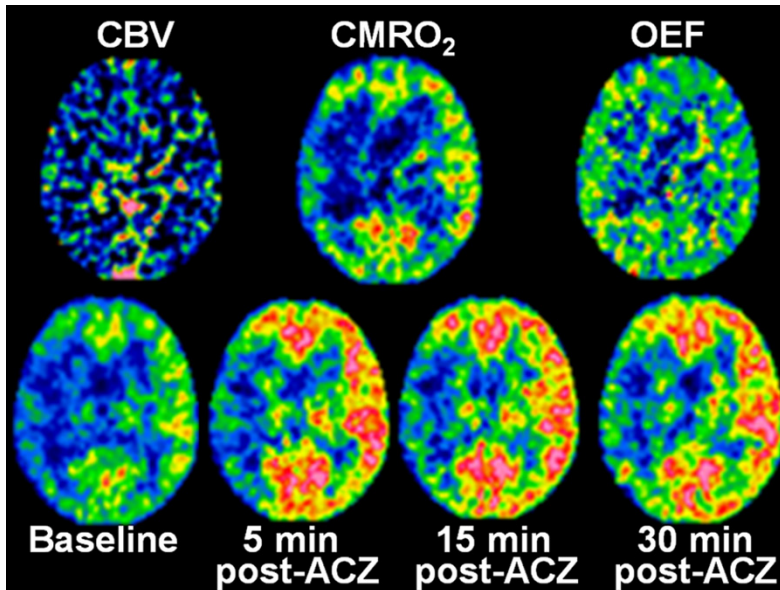


Figure 4. A 65-year-old man with minor stroke of left motor weakness due to right middle cerebral artery (MCA) occlusion and asymptomatic left internal carotid artery stenosis. In the symptomatic right MCA territory, baseline cerebral blood flow (CBF) and cerebral metabolic rate of oxygen ($CMRO_2$) were reduced and cerebral blood volume (CBV) and oxygen extraction fraction (OEF) were normal. In that territory, CBF continued to increase gradually until 30 min after acetazolamide (ACZ) administration. By contrast, in the asymptomatic left MCA territory, CBF was greatly increased at 5 min after ACZ administration and then slightly decreased 25 min later.

those of healthy subjects or patients with normal CBV and $CMRO_2$. An increase in CBV combined with a decrease in CBF implies a long mean transit time (MTT), which is a function of CBV/CBF that can result in poor drug delivery, thereby leading to little or no CVR immediately following ACZ administration in regions affected by such a condition. By contrast, because of normal drug delivery, CVR in surrounding or other brain tissues with a normal MMT shows a fast reaction. Moreover, in regions with increased CBV, the small arterioles or intraparenchymal vessels are dilated. These preexisting dilated vessels are unable to respond immediately after ACZ administration, in contrast to normal vessels in surrounding or other brain tissue, which dilate rapidly. These differences in CVR to ACZ administration can result in a sudden decrease in the cerebral perfusion pressure in normal vessels relative to preexisting dilated vessels, and in turn, this dynamically developed pressure gradient can result in the early “steal” of blood flow from preexisting dilated to normal vessels, after which, the preexisting dilated vessels may gradually respond to ACZ. By contrast, a vasodilation effect of ACZ

may gradually reduce with time in normal vessels. These mechanisms could help explain the paradoxical and transient reduction in CBF immediately after ACZ administration (the early “steal” phenomenon) and its subsequent return.

For MCA territories with abnormally decreased $CMRO_2$, CBF did not change at 5 min after ACZ administration and thereafter, increased and maintained this value until about 30 min after ACZ administration. ACZ acts via carbonic anhydrase, and a reduction in its activity slows CVR to ACZ, and the activity of carbonic anhydrase is thought to depend on cerebral metabolism. In this study, CBF was lower in MCA territories with abnormally decreased $CMRO_2$ than in those of healthy subjects or patients

with normal CBV and $CMRO_2$. Reduced CBF can cause poor drug delivery, and consequently, little or no CVR immediately after ACZ administration in the affected region. On the other hand, CBV was lower in MCA territories with abnormally decreased $CMRO_2$ than in those of healthy subjects or patients with normal CBV and $CMRO_2$. The “steal” of blood flow from shrinking to normal vessels by ACZ administration may therefore be difficult. These hypotheses may explain the delayed response of the CBF increase to ACZ in MCA territories with abnormally decreased $CMRO_2$.

The main finding of the present study was that CBF changes at 5 min after ACZ administration can detect abnormally increased CBV and abnormally decreased $CMRO_2$ more accurately than can CBF changes at 15 or 30 min after ACZ administration. By contrast, the accuracy for detecting abnormally increased OEF did not differ among CBF changes at any time point during ACZ challenge. This finding demonstrated that measuring CBF at 5 min after ACZ administration is the optimal timing for the accurate detection of cerebral hemodynamics

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and metabolism in patients with bilateral atherosclerotic ICA or MCA steno-occlusive diseases. In particular, a transient reduction in CBF at 5 min after ACZ administration (early “steal” phenomenon) strongly indicates abnormally increased CBV. No changes or only a slight increase in CBF at 5 min after ACZ administration also suggests abnormally decreased $CMRO_2$. By contrast, the positive predictive value for the detection of abnormally increased OEF was relatively low (50%-60%) for CBF at any time point during ACZ challenge. Cerebral hemodynamics and metabolism in abnormally increased OEF were classified into abnormally increased CBV and abnormally decreased $CMRO_2$. In these two subgroups, relative CBF at any time point after ACZ administration did not differ between the presence or absence of abnormally increased OEF. This may explain the low positive predictive value for CVR to ACZ to detect abnormally increased OEF.

These findings suggest that 3D measurement of cerebral hemodynamics with repeatable and short-time scanning such as $H_2^{15}O$ PET is suitable for detecting early CBF changes after ACZ administration. However, the relatively high cost and limited availability of PET limits its use in the clinical setting. MRI has been shown to be able to quantify the blood oxygen level-dependent (BOLD) changes in venous structures and/or brain parenchyma induced by differences between oxy- and deoxyhemoglobin in terms of magnetic susceptibility [28-30]. A previous study involving patients with steno-occlusive diseases of major cerebral arteries in whom CVR to ACZ was continuously measured using BOLD imaging with 3T MRI reported that overall, BOLD signals began increasing immediately after ACZ administration and approached a plateau at approximately 8.5 min after administration [31]. However, BOLD signals showed more severely reduced relative reactivity at 6-7 min after the initiation of ACZ infusion in some patients, as well as progressive partial recovery toward a pre-ACZ administration value at 10-11 min. Several studies have shown that arterial spin labeling MRI can also display CBF images before and after ACZ with short-time scanning [32-34]. Investigating the relationship between early CBF changes after ACZ administration on BOLD or arterial spin labeling MRI and preexisting cerebral hemodynamics and metabolism on ^{15}O gas PET would therefore be beneficial.

The present study had several limitations. First, because cerebral hemodynamics are impaired more strongly or frequently in patients with bilateral major cerebral steno-occlusive diseases compared with those with unilateral disease [20], we included only the former patients. Therefore, our findings may not be applicable to patients with unilateral major cerebral steno-occlusive diseases. Second, in the present study, CBF was obtained using the autoradiography method with 90 s of scanning after tracer injection four times so that each mid-scan time was at pre-ACZ administration and 5, 15, and 30 min after the end of ACZ administration. By contrast, Okazawa et al. [15] used 3-min dynamic $H_2^{15}O$ PET scans after ACZ injection. This dynamic scan enables chronological changes in CBF after ACZ administration to be obtained in more detail than our method. Third, we enrolled healthy subjects to obtain normal control values of brain ^{15}O gas PET. However, we did not obtain chronological changes in CBF after ACZ administration on $H_2^{15}O$ PET in healthy subjects. The comparisons of these chronological changes in CBF between healthy subjects and patients may make our findings more robust.

In conclusion, CBF measurement at 5 min after ACZ administration appears to be the optimal timing for the detection of preexisting cerebral hemodynamics and metabolism in patients with bilateral atherosclerotic ICA or MCA steno-occlusive diseases.

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