Carbon Dioxide Reactivity by Consecutive Technetium-99m-HMPAO SPECT in Patients with a Chronically Obstructed Major Cerebral Artery

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In the management of major cerebral artery obstruction, cerebral perfusion reserve is key to introducing cerebral revascularization surgery. The purpose of this study was to evaluate the feasibility of assessing cerebral perfusion reserve by consecutive ^{99m}Tc-hexamethyl-propyleneamine oxime (^{99m}Tc-HMPAO) SPECT with 5% carbon dioxide (CO_2) inhalation. **Methods:** The CO_2 inhalation and consecutive ^{99m}Tc-HMPAO SPECT study was performed on 30 chronic ischemic cerebrovascular disease patients with unilateral major cerebral artery obstruction and on 27 patients without. CO_2 reactivity was expressed as the percent increase of ^{99m}Tc-HMPAO accumulation from the baseline (%Change) and as a constant k' that was the ratio of ^{99m}Tc-HMPAO accumulation per 1 mmHg change of end-tidal CO_2 tension by exponential curve fitting. **Results:** The mean %Change and k' in the middle cerebral artery (MCA) territory on the side without an obstructive lesion or in the cerebellum ranged from 10.0% to 11.1% and from 0.98% to 1.13% per mmHg, respectively. In the MCA territory, an obstructive lesion was noted in 5.9% versus 0.54% per mmHg in the contralateral MCA territory ($p < 0.01$). Eleven of 30 patients with major cerebral artery obstruction revealed significant asymmetry in the k' value between bilateral MCA territories. **Conclusion:** The results showed compromised cerebral perfusion reserve in the obstructed major cerebral artery territory. The present method was proven clinically useful for evaluating cerebral perfusion reserve in patients with unilateral major cerebral artery obstruction.

Key Words: ^{99m}Tc-hexamethyl-propyleneamine oxime; carbon dioxide; cerebral perfusion reserve; single-photon emission computed tomography

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In patients with major cerebral artery obstruction, cerebral perfusion reserve is key to introducing cerebral revascularization surgery such as external to internal cranial bypass or carotid endarterectomy. Routine x-ray computed tomography (XCT), magnetic resonance imaging (MRI) and angiographic evaluation is not always accurate in predicting compromised cerebral perfusion reserve (*1*). For optimal therapy, it is clinically important to assess cerebral perfusion reserve.

Carbon dioxide (CO_2) is a potent vasodilator of cerebral blood vessels, and regional cerebral blood flow (rCBF) reactivity to CO_2 is one of the representative parameters in the assessment of cerebral perfusion reserve. After Kety and Schmidt (*2*) showed that the inhalation of CO_2 caused a marked rise in rCBF, cerebrovascular reactivity to changes in CO_2 levels (CO_2 reactivity) has been thoroughly investigated in various disease conditions and in healthy individuals (*3–9*). However, simple methods with high spatial resolution and relatively low cost remain desirable for daily clinical use.

Technetium-99m-hexamethyl-propyleneamine oxime (^{99m}Tc-HMPAO) (*10*) is a recently developed radiopharmaceutical for use in brain perfusion imaging by single-photon emission computed tomography (SPECT). It is rapidly distributed in proportion to rCBF, crosses the intact blood-brain barrier and is converted into hydrophilic compounds that hardly return to the bloodstream. This process occurs within a few minutes after ^{99m}Tc-HMPAO is intravenously administered, and its retention in the brain is stable for hours (*11*). Therefore, the pattern of its distribution is a "frozen" perfusion image at the time of administration. Thereafter, additional accumulation of ^{99m}Tc-HMPAO into brain tissue should be acquired by subtraction of two consecutive ^{99m}Tc-HMPAO SPECT images. This means that cerebral perfusion images under different conditions such as normocapnic and hypercapnic conditions can be obtained within a short time.

The purpose of this study was to establish the feasibility of assessing cerebral perfusion reserve in patients with unilateral major cerebral artery obstruction by consecutive ^{99m}Tc-HMPAO SPECT with 5% CO_2 inhalation.

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TABLE 1A
Clinical Features and Physiological Data with or Without CO_2 and CO_2 Reactivity in Group NS (No Stenosis)

Patient no.	Sex	Age (yr)	Diagnosis	Blood pressure (Systole/Diastole, mmHg)		Heart rate (beats min^{-1})		P_ECO_2 (mmHg)	
				$CO_{2(-)}$	$CO_{2(+)}$	$CO_{2(-)}$	$CO_{2(+)}$	$CO_{2(-)}$	$CO_{2(+)}$
1	M	70	Dizziness	122/70	137/72	58	64	31	40
2	M	79	TIA	116/66	129/76	81	82	30	39
3	F	69	Dizziness	122/70	142/78	66	71	37	52
4	F	74	Dizziness	120/78	134/82	68	70	46	57
5	M	61	Dizziness	108/76	119/78	79	76	32	47
6	M	58	RIND	110/67	120/69	50	56	40	53
7	M	59	Headache	104/67	116/67	58	66	43	51
8	F	57	Dizziness	145/74	171/74	73	72	42	50
9	F	53	Dizziness	141/72	160/84	66	65	41	50
10	M	74	Comp stroke	111/68	120/69	59	72	33	40
11	M	61	Comp stroke	150/79	164/84	53	64	26	35
12	M	60	Dizziness	125/82	133/94	79	82	30	39
13	F	71	Dizziness	130/76	150/86	61	68	31	40
14	M	40	Dizziness	108/70	118/70	64	64	35	43
15	F	20	Dizziness	99/71	113/77	74	78	29	41
16	F	70	Comp stroke	129/80	134/85	57	62	28	40
17	M	77	Headache	135/78	156/80	58	64	22	34
18	M	50	Comp stroke	152/74	174/98	79	85	27	35
19	M	63	RIND	113/65	133/74	74	82	40	50
20	M	66	Comp stroke	130/76	154/86	75	80	35	41
21	M	52	Comp stroke	120/77	132/83	71	75	36	46
22	M	57	Comp stroke	146/74	156/86	78	82	41	49
23	M	57	Comp stroke	118/67	134/72	59	62	30	39
24	M	67	Dizziness	142/78	152/84	60	68	36	43
25	M	24	Headache	112/67	121/68	74	82	41	46
26	M	75	Comp stroke	108/68	116/74	58	64	39	51
27	M	67	Headache	150/85	166/92	64	72	37	45
		60.4 ± 14.2		125 ± 16/73 ± 5	139 ± 19*/ 79 ± 8*	67 ± 9	71 ± 8*	34.7 ± 6.0	44.3 ± 6.1*

See Table 1B for definitions.

METHODS

Subjects

We assessed the reproducibility between two consecutive ^{99m}Tc-HMPAO SPECT without CO_2 challenge in a baseline study of 14 patients (group NC; no CO_2 challenge, mean age 51.0 yr) with various central nervous system disorders.

Cerebral perfusion reserve was assessed in another series of 57 patients (45 males and 12 females) varying from 20 to 79 yr (58.8 ± 12.7 yr; mean ± s.d.). They were all in the chronic stage of ischemic cerebrovascular disease (CVD) or suffering from non-specific brain symptoms without focal signs (i.e., headache, dizziness and syncope). All of them had XCT examinations and had undergone cerebral angiography either by conventional technique or by digital subtraction angiography. They had no cerebellar involvement on neurological and XCT examination. No patient had a cerebral infarction larger than 3 cm in the long-axis or hemorrhagic infarction on XCT examination. All patients were alert and had no cardiopulmonary complications. The SPECT study was performed at least 4 wk after the last episode.

As presented in Tables 1 and 2, the latter series of patients were classified into two groups (Group NS; 27 patients without stenosis, Group OB; 30 patients with unilateral obstruction) according to cerebral angiography results. On angiograms, patients in Group NS had no stenotic lesions in the internal carotid artery (ICA) or in the main trunk of middle cerebral artery (MCA). In Group OB, as listed in Table 2, 11 patients had severe ICA stenosis, 8 had ICA occlusion, 5 had severe MCA stenosis and 6 had MCA occlusion. All lesions were located on the unilateral side. Severe stenosis was diagnosed when luminal narrowing of over 75% in diameter was observed on angiograms. There was no significant difference between the two groups in age distribution. Stroke patients (complete stroke, transient ischemic attack and reversible ischemic neurological deficit) were more predominant in Group OB than those in Group NS (90% versus 44%; $p < 0.01$ by chi-square test). On XCT, 11 of 27 patients in Group NS had 11 ischemic lesions, one of which was located in the cerebral cortex and the others were located in the subcortex. In Group OB, there were 22 cortical and 13 subcortical ischemic lesions in 19 cerebral hemispheres on the side of an obstructive lesion (affected side), while there were two cortical and five subcortical ischemic lesions in six contralateral hemispheres (non-affected side). In Group OB, there was a higher incidence of cortical involvement on the affected side compared with that in Group NS ($p < 0.01$ by chi-square test).

The study was performed under the standard ethical guidelines of Osaka University and informed consent was obtained from each individual prior to the study.

TABLE 1B
HMPAO SPECT Results for Group NS

Patient no.	%Change			k′ (%mmHg^{-1})			
	R MCA	L MCA	Cerebellum	R MCA	L MCA	Cerebellum	Δk′
1	5.5	9.2	6.0	0.61	0.98	0.65	0.38
2	6.8	7.7	19.3	0.74	0.84	1.96	0.10
3	11.3	14.9	20.9	0.71	0.93	1.26	0.21
4	7.6	9.1	8.4	0.66	0.79	0.74	0.13
5	8.3	7.8	7.1	0.54	0.51	0.45	−0.03
6	2.9	3.0	4.4	0.21	0.22	0.34	0.01
7	32.9	28.4	22.9	3.56	3.14	2.60	−0.42
8	7.9	10.6	2.6	0.94	1.26	0.33	0.32
9	4.2	4.5	7.8	0.46	0.48	0.84	0.02
10	16.5	14.1	12.9	2.20	1.90	1.72	−0.31
11	8.1	8.4	5.2	0.85	0.88	0.56	0.03
12	14.0	11.8	6.8	1.45	1.23	0.75	−0.22
13	16.9	13.2	14.0	1.75	1.36	1.45	−0.39
14	22.3	18.3	14.9	2.50	2.10	1.75	−0.40
15	20.3	20.7	7.1	1.54	1.57	0.57	0.03
16	12.0	12.1	11.1	0.95	0.95	0.90	0.00
17	6.7	7.5	10.2	0.53	0.62	0.80	0.08
18	15.8	21.0	22.0	1.81	2.38	2.46	0.57†
19	13.1	12.8	14.2	1.24	1.21	1.33	−0.03
20	−4.7	−6.8	5.6	−0.78	−1.18	−0.97	−0.39
21	11.0	5.8	18.2	1.06	0.59	1.67	−0.47
22	26.9	26.4	25.1	2.99	2.92	2.78	−0.07
23	8.8	7.1	9.3	0.94	0.75	0.98	−0.19
24	11.6	9.7	6.7	1.56	1.32	0.92	−0.25
25	−0.2	0.9	5.8	−0.05	0.19	1.11	0.24
26	−0.2	−0.6	1.6	−0.02	−0.06	0.14	−0.04
27	12.4	11.7	16.6	1.46	1.40	1.91	−0.07
	11.1 ± 8.2	10.8 ± 7.6	10.9 ± 7.3	1.13 ± 0.94	1.08 ± 0.89	1.11 ± 0.83	−0.04 ± 0.26

P_ECO_2 = end-tidal carbon dioxide tension; %Change = percent increase of ^{99m}Tc-HMPAO accumulation by carbon dioxide inhalation from baseline; k′ = the ratio of ^{99m}Tc-HMPAO accumulation per 1 mmHg change of P_ECO_2 by exponential curve fitting, $CO_{2(+)}$ and $CO_{2(-)}$ are conditions with and without carbon dioxide inhalation respectively, R = right; L = left; Δk′ = difference of k′ values between bilateral MCA (L-R); TIA = transient ischemic attack; RIND = reversible ischemic neuronal deficit; Comp stroke = complete stroke.

The bottom data line presents mean ± s.d.; *: $p < 0.01$ vs. $CO_{2(-)}$ by paired t-test; †significantly asymmetric Δk′.

Device and Study Protocol

We used a high-performance, four-head rotating gamma camera equipped with low-energy, general-purpose, parallel-hole collimators (*12*) (Gamma View SPECT 2000H, Hitachi Medical Co., Japan). The spatial resolution was 13.0 mm FWHM. SPECT acquisition was performed in 64 steps, 360° and with a 64 × 64 matrix (4 × 4 mm per pixel).

The patients were rested supine with their eyes closed, and their heads were affixed with gummed tape to a ready-made semicylindrical plastic holder by the forehead and the chin. The space between the temporal head and the holder was filled with a soft rubber sponge. The headholder was adjusted with the aid of positioning a light beam so that the orbitomeatal line was perpendicular to the axis of camera rotation. A temporal profile of end-tidal CO_2 tension (P_ECO_2) was continuously monitored in the nasal cavity by an infrared CO_2 analyzer (Respina IH26, Nihon San-ei, Japan) during the procedure. Systemic blood pressure and heart rate were monitored using an automatic sphygmomanometer. The study protocol is shown in Figure 1. Patients in Group NS or OB at first inhaled 5% CO_2 gas (5% CO_2 in 20% oxygen and 75% nitrogen gas) through a naso-oral mask. When P_ECO_2 reached a plateau, 185 MBq of ^{99m}Tc-HMPAO (Ceretec®, Amersham, Japan) was injected into a cubital vein via a butterfly needle. Five minutes of further 5% CO_2 inhalation after injection was followed by the first SPECT study for 15 sec per step. On the other hand, patients in Group NC breathed room air during this phase of the procedure. All patients breathed room air after the first SPECT study had started. Immediately after this, without repositioning the patient and using the same vial as in the first injection, 740 MBq of ^{99m}Tc-HMPAO was injected. Five minutes after the second injection, the second SPECT study was begun for 5 sec per step. Residual syringe radioactivity was measured after each injection to correct the actual amount of radioactivity administered. The total examination time was approximately 35 min.

Image Processing and Region of Interest (ROI) Analysis

SPECT acquisition data sets were prefiltered with a Butterworth filter then reconstructed with a Ramachandran backprojection filter. Chang's postreconstruction attenuation correction was applied with an attenuation coefficient of 0.08 cm^{-1} to the transaxial image data. The final reconstructed transaxial image was 8 mm thick per slice.

The cerebral perfusion image produced by the second ^{99m}Tc-HMPAO study (I_2') was acquired as follows:

TABLE 2A

Clinical Features and Physiological Data with or Without CO_2 and CO_2 Reactivity in Group OB (Unilateral Obstruction)

Patient no.	Sex	Age (yr)	Diagnosis	Vascular lesions	Blood pressure (Systole/Diastole, mmHg)		Heart rate (beats min^{-1})	
					$CO_{2(-)}$	$CO_{2(+)}$	$CO_{2(-)}$	$CO_{2(+)}$
1	M	59	TIA	R ICA occlusion	128/76	130/80	75	78
2	M	43	Comp stroke	L ICA severe stenosis	118/77	126/82	81	84
3	M	50	Comp stroke	R ICA severe stenosis	149/92	160/97	86	88
4	M	66	Comp stroke	R ICA severe stenosis	119/81	142/85	78	80
5	M	75	Comp stroke	L ICA severe stenosis	142/74	152/83	100	98
6	M	67	TIA	L ICA severe stenosis	104/69	116/75	90	92
7	M	48	TIA	R MCA severe stenosis	161/103	148/103	72	70
8	M	52	Comp stroke	L MCA severe stenosis	128/84	134/88	84	87
9	M	35	TIA	L MCA severe stenosis	117/83	124/80	63	70
10	F	65	Comp stroke	L MCA occlusion	150/76	151/66	74	78
11	M	63	Comp stroke	R ICA occlusion	96/65	105/68	54	60
12	M	68	TIA	R ICA severe stenosis	104/70	116/70	59	63
13	F	67	TIA	L MCA severe stenosis	100/66	120/68	55	62
14	F	49	Comp stroke	L ICA occlusion	107/73	116/72	58	62
15	F	70	TIA	L ICA occlusion	150/95	160/96	67	73
16	M	51	Comp stroke	L MCA occlusion	110/72	120/74	71	76
17	M	40	TIA	R ICA severe stenosis	143/82	150/88	64	66
18	M	57	Comp stroke	R ICA occlusion	99/66	116/66	74	78
19	M	59	Comp stroke	R MCA occlusion	130/86	140/96	80	84
20	M	54	Syncope	R MCA occlusion	149/87	156/90	68	72
21	M	67	Comp stroke	R ICA severe stenosis	131/70	140/73	55	61
22	M	59	TIA	R ICA severe stenosis	110/74	121/82	80	82
23	F	65	Dizziness	R ICA severe stenosis	140/85	146/88	58	63
24	M	76	Comp stroke	L ICA severe stenosis	148/92	152/92	79	81
25	M	71	Headache	R ICA occlusion	102/68	109/71	65	74
26	M	57	Comp stroke	R ICA occlusion	120/70	130/76	56	61
27	M	55	Comp stroke	L MCA occlusion	103/69	118/70	56	60
28	M	29	TIA	L MCA occlusion	130/91	136/96	79	84
29	M	49	Comp stroke	R MCA severe stenosis	108/71	126/77	80	83
30	M	58	TIA	L ICA occlusion	106/68	123/71	55	59
		57.4 ± 11.5			123 ± 19/78 ± 10	133 ± 16*/ 81 ± 11*	71 ± 12	74 ± 11*

See Table 2B for definitions.

$$I_2' = I_2 \times (15\ sec/5\ sec) - I_1',$$

where I_2 and I_1' are the second and decay-corrected initial SPECT images reconstructed, respectively. Two sets of transaxial perfusion images produced by the first and the second ^{99m}Tc-HMPAO studies were normalized by the administered ^{99m}Tc-HMPAO radioactivity.

ROI analysis in this study is illustrated in Figure 2. In transaxial images produced by the first and second ^{99m}Tc-HMPAO studies, the corresponding sets of six consecutive slices from the basal ganglia and the thalamic level to the parietal cortex level were selected so that they included most of the MCA territories. Among these, the top three and bottom three slices were added separately to create two integrated images that were outlined by the 55% count cutoff to maximum ^{99m}Tc-HMPAO accumulation to trace the contour of the cerebrum. The outlined cerebral area was automatically divided into three equal sized longitudinal ROIs on the top integrated image and into four ROIs on the bottom integrated image. Bilateral outer ROIs on each integrated image corresponded to bilateral MCA territories. Two or three slices including the cerebellum were also added and outlined by the 65% count cutoff. Technetium-99m-HMPAO accumulation in these MCA and cerebellar ROIs were summed over the images and averaged by pixel and slice. For semiquantitative analysis, the %Change and the CO_2 reactivity constant k' value [Olesen et al. (*13*)] were calculated in each territory as follows:

$$\%Change = 100 \times \frac{HMPAO_{1st} - HMPAO_{2nd}}{HMPAO_{2nd}}$$

$$k' = 100 \times \frac{\ln (HMPAO_{1st}) - \ln (HMPAO_{2nd})}{\text{change of } P_ECO_2},$$

where $HMPAO_{1st}$ and $HMPAO_{2nd}$ were the first and the second ^{99m}Tc-HMPAO accumulations in a corresponding ROI, respectively. The difference in the k' value between bilateral MCA territories was expressed as $\Delta k'$ by subtracting k' in the affected MCA (or right MCA in Group NS) from that in the nonaffected MCA (or left MCA in Group NS).

The ROI data were analyzed by paired or unpaired Student's t-test. A difference of $p < 0.05$ was considered significant.

TABLE 2B
HMPAO SPECT Results for Group OB

Patient no.	P_ECO_2 (mmHg)		%Change (%)			k′ (% $mmHg^{-1}$)			
	$CO_{2(-)}$	$CO_{2(+)}$	NA MCA	AF MCA	Cerebellum	NA MCA	AF MCA	Cerebellum	Δk′
1	40	50	16.9	15.8	17.4	1.57	1.46	1.59	0.11
2	40	50	13.4	12.2	14.5	1.27	1.15	1.34	0.12
3	37	48	13.0	4.3	11.9	1.10	0.39	1.02	0.72**
4	31	47	13.0	9.0	15.0	0.76	0.54	0.87	0.22
5	36	47	10.8	12.2	16.0	0.95	1.05	1.36	−0.10
6	42	49	20.8	16.3	20.7	2.72	2.16	2.68	0.56**
7	37	47	26.0	22.7	26.8	2.32	2.06	2.38	0.26
8	36	44	15.8	13.4	11.4	1.83	1.59	1.36	0.24
9	40	50	22.1	23.0	27.0	2.00	2.08	2.38	−0.08
10	35	42	16.5	2.4	20.7	2.17	0.35	2.70	1.82**
11	37	45	11.5	−6.6	16.1	1.35	−0.86	1.88	2.21**
12	34	41	10.8	10.2	17.0	1.47	1.39	2.24	0.08
13	31	37	−1.5	−3.6	−1.8	−0.25	−0.63	−0.29	0.38
14	27	40	9.3	1.8	3.9	0.68	0.15	0.29	0.53**
15	33	43	5.0	−2.2	12.7	0.49	−0.22	1.20	0.71**
16	24	37	19.5	20.6	24.6	1.37	1.44	1.69	−0.07
17	33	44	−9.4	−9.8	−7.8	−0.89	−0.95	−0.74	0.05
18	28	33	1.1	−3.4	2.1	0.21	−0.68	0.42	0.89**
19	40	50	12.3	10.2	12.6	1.15	0.96	1.19	0.19
20	40	50	6.1	9.2	7.4	0.57	0.89	0.72	−0.32
21	32	40	16.8	6.0	16.0	1.95	0.73	1.87	1.23**
22	37	47	10.1	0.5	3.8	0.95	0.06	0.38	0.89**
23	33	40	−1.6	−1.0	−2.0	−0.25	−0.13	−0.30	−0.11
24	32	40	14.0	12.0	13.7	1.65	1.39	1.60	0.26
25	35	43	−0.2	−1.3	0.2	−0.04	−0.17	0.02	0.13
26	25	40	−1.7	−12.9	−2.2	−0.11	−0.92	−0.14	0.80**
27	30	44	8.1	7.6	14.0	0.56	0.52	0.94	0.04
28	43	53	4.2	1.8	5.3	0.40	0.17	0.51	0.23
29	37	50	5.1	−0.3	1.5	0.38	0.13	0.11	0.26
30	42	55	13.5	4.0	10.8	0.99	0.23	0.80	0.69**
	34.9 ± 5.0	44.9 ± 5.2*	10.0 ± 8.1	5.9 ± 9.2†‡	11.0 ± 9.1	0.98 ± 0.85	0.54 ± 0.91†‡	1.07 ± 0.93	0.43 ± 0.56§

P_ECO_2 = end-tidal carbon dioxide tension, %Change = percent increase of ^{99m}Tc-HMPAO accumulation by carbon dioxide inhalation from baseline; k′ = the ratio of ^{99m}Tc-HMPAO accumulation per 1 mmHg change of P_ECO_2 by exponential curve fitting; $CO_{2(+)}$ and $CO_{2(-)}$ are conditions with and without carbon dioxide inhalation respectively; R = right, L = left; NA = nonaffected side, AF = affected side; Δk′ = difference of k′ values between bilateral MCA (NA-AF); TIA = transient ischemic attack; Comp stroke = complete stroke.

The bottom data line presents mean ± s.d.; *$p < 0.01$ vs. $CO_{2(-)}$ by paired t-test; †$p < 0.05$ vs. R MCA, L MCA and cerebellum in Group NS by unpaired t-test; ‡$p < 0.01$ vs. NA MCA and cerebellum in Group OB by paired t-test; §$p < 0.01$ vs. group NS by unpaired t-test; **significantly asymmetric Δk′.

Statistical Error by Subtraction Method

When two consecutive SPECT acquisitions are performed under the same conditions and if ^{99m}Tc-HMPAO accumulations are proportional to the dose administered, the following equation should hold:

$$C_2 \fallingdotseq R_t \times C_1 + R_d \times R_t \times C_1,$$

where C_1 and C_2 are observed counts in a ROI on the first and second original SPECT images, R_t and R_d are the acquisition time ratio and the administered ^{99m}Tc-HMPAO dose ratio of the second SPECT against the first, respectively. Acquisition time-corrected C_2 (C_2') and the count after subtraction (C_{subt}) are calculated using the following equations.

$$C_2' = C_2 \times R_t^{-1}$$

$$C_{subt} = C_2' - C_1.$$

Because C_1 and C_2 are independent events with Poisson's fluctuation, the statistical variance after subtraction (var (C_{subt})) is equal to the addition of var (C_1) and var (C_2') as follows:

$$\text{var}\,(C_{subt}) = C_1 + \frac{C_2}{R_t^2}.$$

After normalization by the dose ratio (R_d), the final statistical variance after subtraction (var (C_{subt}')) is as follows:

$$\text{var}\,(C_{subt}') = \frac{1}{R_d^2} \times \left(C_1 + \frac{C_2}{R_t^2}\right)$$

$$\fallingdotseq \frac{1}{R_d^2} \times \left(1 + \frac{1 + R_d}{R_t}\right) \times C_1$$

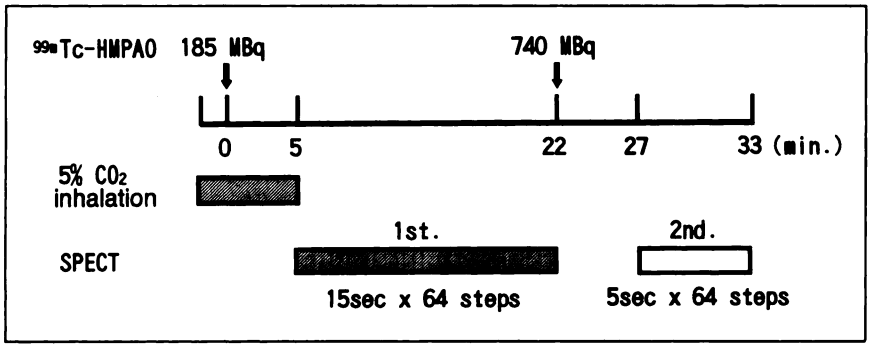

FIGURE 1. Protocol for consecutive ^{99m}Tc-HMPAO SPECT with CO_2 inhalation. Patients inhale 5% CO_2 at the beginning of the study and two consecutive SPECT studies are performed. Hatched boxes are duration of CO_2 inhalation (upper) and SPECT image acquisition corresponding to hypercapnic condition (lower). Unfilled box is SPECT image acquisition corresponding to normocapnic conditions, including imaging under hypercapnia. Elapsed time includes camera rotation time during SPECT imaging.

$$\fallingdotseq \frac{1}{R_d^2} \times \left(1 + \frac{1 + R_d}{R_t}\right) \times \text{var}\,(C_1).$$

Figure 3 shows the relationships among the ratio of var (C'_{subt}) against var (C_1), R_t and R_d. In the present study, we used $R_d = 4$ and $R_t = 1/3$, then var (C'_{subt}) = var (C_1).

RESULTS

Reproducibility Between the First and the Second ^{99m}Tc-HMPAO Study at Rest

Figure 4 shows relationship between the first and the second mean ^{99m}Tc-HMPAO accumulation in bilateral MCA territories and the cerebellum (total 42 ROI) in Group NC. There was good agreement between the first and the second ^{99m}Tc-HMPAO accumulations. The %Change of this condition was 0.2 ± 6.0.

CO_2 Challenge Test

CO_2 inhalation caused no apparent side effects except for mild tachypnea. All patients underwent this challenge with safety. The individual physiological parameters with and without CO_2 inhalation are shown in Tables 1 and 2. There was significant rise in P_ECO_2, systolic and diastolic blood pressure and heart rate during CO_2 inhalation from those without CO_2. There was no significant difference between Groups NS and OB in these parameters.

Tables 1 and 2 also show the individual results of %Change, k′ and Δk′ in each region. CO_2 challenge caused significant increase of ^{99m}Tc-HMPAO accumulation in all regions in Groups NS and OB compared with that in Group NC. Outside the affected MCA territory (AF MCA) in Group OB, %Change by CO_2 challenge ranged from 10% to 11.1% by the mean value. In the affected MCA, both %Change (5.9% ± 9.2%) and k′ (0.54 ± 0.91 %/mmHg) were significantly lower than those in the nonaffected MCA territory (NA MCA), than those in the cerebellum in Group OB or than those in all territories in Group NS. There was no significant difference between Group NS and Group OB in cerebellar CO_2 reactivity.

In Group NS, there was no correlation ($r = -0.15 \sim -0.06$, $p = 0.45 \sim 0.74$) among age and the k′ values in all territories. And there was no significant difference ($p = 0.50 \sim 0.80$ by unpaired t test) between stroke and nonstroke patients in the mean k′ values. There was correlation ($r = 0.80$ and 0.82, $p < 0.01$) between k′ value in the cerebellum and those in bilateral MCA territories in this group.

As shown in Tables 1 and 2, patients in Group OB had a significantly larger mean Δk′ value than those in Group NS (0.43 ± 0.56 %/mmHg versus −0.04 ± 0.26 %/mmHg; $p < 0.01$). According to the distribution of Δk′ in Group NS patients, the range of significant asymmetry is above 0.48 %/mmHg (mean + 2 s.d.). With this criterion, 11 of 30 patients in Group OB revealed significant asymmetry in CO_2 reactivity between bilateral MCA territories in comparison to 1 of 27 patients in Group NS. Six of eight ICA occlusive patients, four of eleven ICA stenotic patients and one of six MCA occlusive patients showed significant asymmetry.

Figure 5 illustrates a patient (no. 11 in Table 2) with right ICA occlusion. The patient had mild fixed left hemiplegia due to old cerebral infarction and had experienced transient, repeated worsening of the symptom. Consecutive ^{99m}Tc-HMPAO SPECT with 5% CO_2 inhalation (Fig. 5A) demonstrated impaired CO_2 reactivity in the right MCA

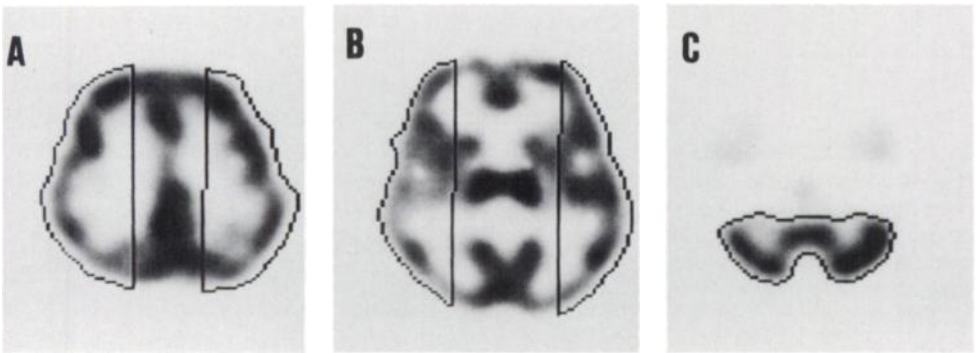

FIGURE 2. ROI analysis. (A and B) Automatically drawn ROIs which correspond to bilateral middle cerebral artery territories at the parietal lobe and the thalamic level, respectively. (C) Automatically drawn ROI which corresponds to the cerebellum.

territory ($\Delta k'$ = 2.21 %/mmHg). After superficial temporal artery to middle cerebral artery bypass surgery, he had no relapse of worsening of the symptom. The follow-up SPECT study (Fig. 5B) revealed improvement of asymmetry in CO_2 reactivity between bilateral MCA territories ($\Delta k'$ = −0.13 %/mmHg).

DISCUSSION

The advantage of the present method is that brain perfusion images with high spatial resolution under various conditions can be obtained within 35 min. Rapid tracer accumulation in the brain allows paradigm or challenge to be continued only for a few minutes. With this imaging technique and CO_2 inhalation, we semiquantified impaired CO_2 reactivity in affected MCA territories.

Other brain perfusion imaging radiopharmaceuticals for SPECT such as ^{133}Xe and N-isopropyl p-[^{123}I] iodoamphetamine (^{123}I-IMP) have been used to assess CO_2 reactivity. Xenon-133 SPECT provides quantitative rCBF measurement repeatedly under various conditions (*7,14–16*). This technique, however, requires a highly sensitive detection system and the acquired image has poor spatial resolution. Iodine-123-IMP SPECT is used for quantitative rCBF measurement (*17*). Unfortunately, due to its long half-life and prolonged accumulation in the brain, two separate SPECT studies with relatively long intervals are required.

On the other hand, positron emission tomography (PET) is a powerful tool for assessing CVD (*5,6,18–20*). It provides quantitative information about rCBF, metabolism and other physiological parameters such as the oxygen extraction fraction. A cyclotron-based PET system is currently too expensive for daily clinical use.

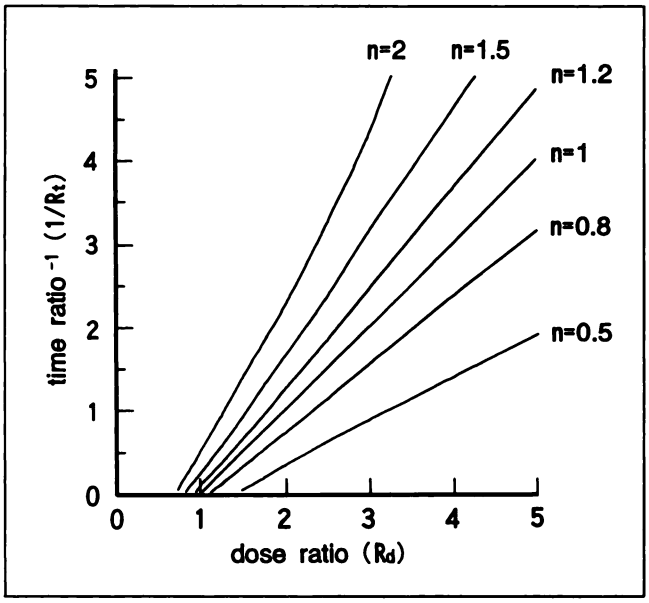

FIGURE 3. Statistical variance of the subtracted count in various combinations of the time and dose ratio between the two SPECT studies. n = var (C'_{subt})/var (C_1) is the ratio of statistical variance in a corresponding ROI on the subtracted image against that on the first SPECT image. R_d and R_t are the administered dose and acquisition time ratios, respectively, of the second study SPECT against the first.

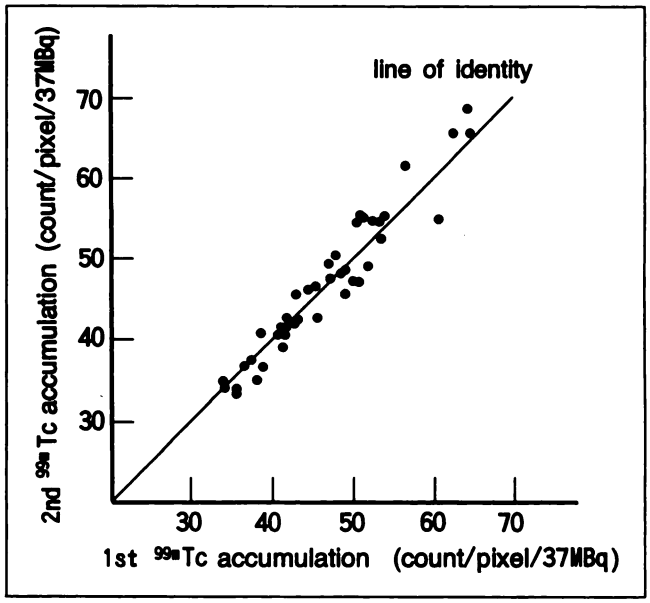

FIGURE 4. Comparison of mean ^{99m}Tc-HMPAO accumulation produced by first and the second administrations of tracer. The equation of the regression line is y = 1.09x − 3.9; the correlation coefficient is 0.95, which is significant ($p < 0.01$).

In the present study, ^{99m}Tc-HMPAO accumulation with 5% CO_2 inhalation increased approximately 11% in mean value from the baseline in Group NS, whereas investigations using other methods have reported that CBF increased from 17% to 68% in the nonaffected hemisphere with the same stimulation (*6,14,21*). The major disadvantage of ^{99m}Tc-HMPAO is its high backdiffusion in the early phase, especially in high flow areas. Its accumulation is no more linear to true rCBF in high flow rates (*11,22*). Thus, true rCBF response to CO_2 was underestimated.

Because the consecutive SPECT and subtraction technique encounters subtraction errors, the values of R_d and R_t must be carefully selected. The larger these values, the smaller the subtraction error (Fig. 3). When patient motion occurs between the two SPECT studies, a large R_d will help salvage the rest image by visual assessment without subtraction because it diminishes the contamination of the first SPECT into the second.

On the other hand, too large R_d results in longer time intervals between tracer injections. If both the first and the second injection are prepared from the same ^{99m}Tc-HMPAO vial, there is a limit in the time between injections. Neirinckx et al. (*10*) recommended that the tracer should be used within 30 min of reconstitution, and Ballinger et al. (*23*) reported that it should be injected within 20 min, in consideration of the deterioration in radiochemical purity. Although our procedure does not precisely satisfy the latter recommendation, good reproducibility of two ^{99m}Tc-HMPAO studies indicates that radiochemical purity is maintained at a satisfactory level to obtain high quality

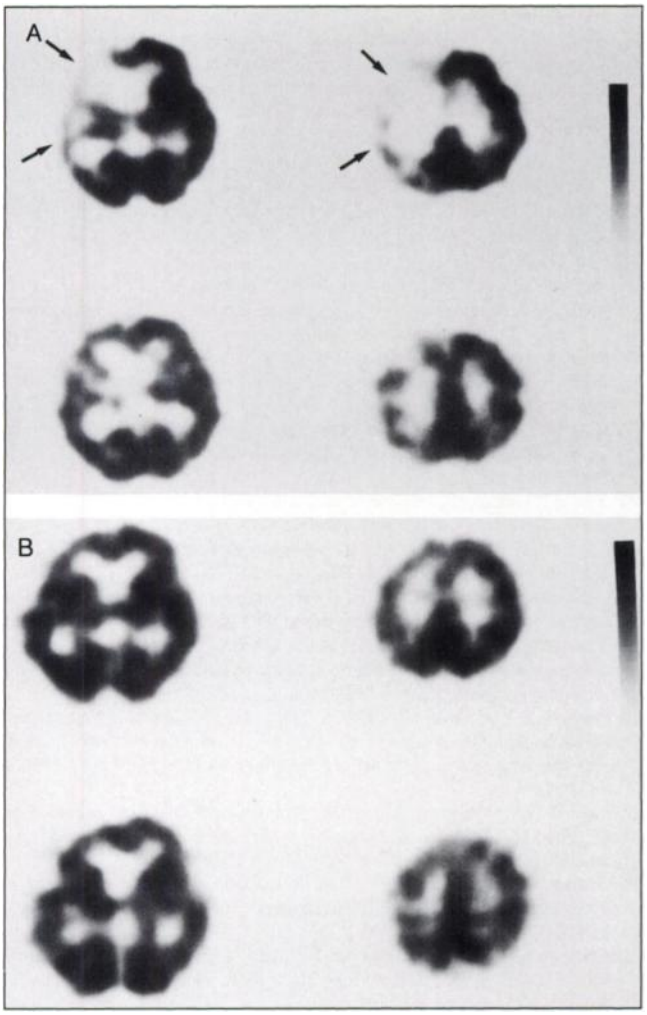

FIGURE 5. Consecutive ^{99m}Tc-HMPAO SPECT studies performed on a patient (a 63-yr-old male) with right ICA occlusion before (A) and after (B) external to internal cranial bypass surgery. Preoperative SPECT study (A) demonstrated mild hypoperfusion during normocapnia and poor response of rCBF to CO_2 in the right MCA territory (arrows) when compared with the left. A postoperative follow-up SPECT study (B) demonstrated improvement in impaired CO_2 reactivity in the right MCA territory. Upper row: brain perfusion images during hypercapnia; lower row: those during normocapnia.

images. Based on these considerations, we selected $R_d = 4$ and $R_t = 1/3$.

Choksey et al. (*24*) have reported that CO_2 inhalation produced no significant increase in ^{99m}Tc-HMPAO accumulation in eight patients. As shown in Tables 1 and 2 and in previous reports (*6,25*), CO_2 reactivity values varied widely, and small numbers of patients might fail to show a statistical difference. The distribution of %Change and k′ values widely overlapped among regions or groups. These parameters alone could not predict compromised cerebral perfusion reserve in individual assessments. Individual assessment of cerebral perfusion reserve must take into account other regions such as the contralateral MCA territory or the cerebellum.

As a stress test for CBF, acetazolamide is also available. Administration of acetazolamide at a dose of 1000 mg also causes submaximum dilatation the of cerebral small vessels (*26*). It is now frequently used for assessment of cerebral vasomotor reactivity because it has little effect on systemic physiological parameters. Although CO_2 inhalation technique has some problems and limits for regular use (*27*), this conventional stress test has been thoroughly analyzed and is still as reliable as the acetazolamide challenge test (*28*). Furthermore, the ratio of detected asymmetric cerebral vasomotor reactivity against Group OB in this present study was comparable to recent studies using ^{99m}Tc-HMPAO and acetazolamide (*29*).

Because age-matched healthy volunteers were not available, we included symptomatic patients in Group NS as references. Although patients with symptomatic ischemic episodes are known to show limited cerebrovascular reactivity (*3,26*), there was no significant difference in mean k′ values between stroke and nonstroke patients in the present study. As shown in a previous study using transcranial Doppler method (*3*), our study shows that major artery obstruction was a more potent attenuater of cerebrovascular reactivity than microangiopathy.

Our results demonstrated impaired CO_2 reactivity in the affected MCA territory in patients with unilateral major cerebral artery obstruction. In patients with obstructed major cerebral arteries, cerebral revascularization surgery is an effective strategy to reduce the risk of CVD (*30,31*). Furthermore, patients with compromised cerebral perfusion reserve will benefit by cerebral revascularization surgery. Due to wide normal ranges as noted above, a reduced k′ value itself does not instantly identify compromised cerebral perfusion reserve in an individual. Some previous cerebrovascular reactivity studies (*29,32*) also had this problem. The use of hemispheric reactivity differences takes advantage of the structural symmetry of the cerebrum to improve sensitivity for identifying abnormalities. This technique, however, limits one hemisphere to be classified as normal.

In the present study, we selected only patients with unilateral major cerebral artery obstruction for Group OB. The results that cerebellar CO_2 reactivity in Group OB was maintained at the same level as that in Group NS and that CO_2 reactivity in MCA territories showed correlation with that in the cerebellum in Group NS may support the feasibility of assessing cerebral perfusion reserve in patients with bilateral major cerebral artery obstruction using cerebellar CO_2 reactivity as a reference (*33*). In this study, 11 of 30 patients in Group OB showed significant asymmetry in k′ values between bilateral MCA territories. Considering the underestimation of the true rCBF response, only patients with severe and global asymmetry in true CO_2 reactivity between bilateral MCA territories may express an increased $\Delta k'$ value. This means that patients with increased $\Delta k'$ values corresponding to unilaterally compromised cerebral perfusion reserve may fit the indication of cerebral revascularization surgery. Patients without an increased $\Delta k'$ in Group OB were interpreted as follows: varying degree but not severely asymmetric CO_2 reactivity

that ^{99m}Tc-HMPAO could not detect, acquired adequate collateral circulation or intact vasomotor capacity with a matched decline of perfusion to metabolic demand. Further prospective or postoperative studies are required to clarify these issues.

CONCLUSION

In conclusion, consecutive ^{99m}Tc-HMPAO SPECT with 5% CO_2 inhalation is a simple and feasible method for assessing CO_2 reactivity. This method provides semiquantitative data about cerebral perfusion reserve. When combined with conventional neuroimaging techniques such as XCT or angiography, it will contribute greatly to patient management on such issues as: introduction of cerebral revascularization therapy, evaluation of therapeutic effects and follow-up studies in patients with unilateral major cerebral artery obstruction.

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REFERENCES

1. Latchaw RE, Ausman JI, Lee MC. Superficial temporal-middle cerebral artery bypass. A detailed analysis of multiple pre- and postoperative angiograms in 40 consecutive patients. *J Neurosurg* 1979;51:455–465.
2. Kety SS, Schmidt CF. The effects of altered arterial tension of carbon dioxide and oxygen on the cerebral blood flow and carbon dioxide consumption of normal young men. *J Clin Invest* 1948;27:484–492.
3. Maeda H, Matsumoto M, Handa N, et al. Reactivity of cerebral blood flow to carbon dioxide in various types of ischemic cerebrovascular disease: evaluation by the transcranial doppler method. *Stroke* 1993;24:670–675.
4. Ogawa S, Handa N, Matsumoto M, et al. Carbondioxide reactivity of the blood flow in human basilar artery estimated by the transcranial Doppler method in normal men: a comparison with that of the middle cerebral artery. *Ultrasound Med Biol* 1988;14:479–483.
5. Levine RL, Dobkin JA, Rozental JM, Satter MR, Nickles RJ. Blood flow reactivity to hypercapnia in strictly unilateral carotid disease: preliminary results. *J Neurol Neurosurg Psychiatry* 1991;54:204–209.
6. Levine RL, Sunderland JJ, Lagreze HL, Nickles RJ, Rowe BR, Turski PA. Cerebral perfusion reserve indexes determined by fluoromethane positron emission scanning. *Stroke* 1988;19:19–27.
7. Tsuda Y, Hartmann A. Changes in hyperfrontality of cerebral blood flow and carbon dioxide reactivity with age. *Stroke* 1989;20:1667–1673.
8. Merrick MV, Ferrington CM, Cowen SJ. Parametric imaging of cerebral vascular reserves. 1. Theory, validation and normal values. *Eur J Nucl Med* 1991;18:171–177.
9. Wei EP, Seelig JM, Kontos HA. Comparative responses of cerebellar and cerebral arterioles to changes in $PaCO_2$ in cats. *Am J Physiol* 1984;246: H386–388.
10. Neirinckx RD, Canning LR, Piper IM, et al. Tc-99m d,l-HM-PAO: a new radiopharmaceutical for SPECT imaging of regional cerebral blood perfusion. *J Nucl Med* 1987;28:191–202.
11. Andersen AR, Friberg HH, Schmidt JF, Hasselbalch SG. Quantitative measurements of cerebral blood flow using SPECT and [^{99m}Tc]-d,l-HM-PAO compared to Xe-133. *J Cereb Blood Flow Metab* 1988;8:S69–81.
12. Kimura K, Hashikawa K, Etani H, et al. A new apparatus for brain imaging: four head rotating gamma camera single photon emission computed tomograph. *J Nucl Med* 1990;31:603–609.
13. Olesen J, Paulson OB, Lassen NA. Regional cerebral blood flow in man determined by the initial slope of the clearance of intra-arterially injected ^{133}Xe. *Stroke* 1971;2:519–540.
14. Keyeux A, Laterre C, Beckers C. Resting and hypercapnic rCBF in patients with unilateral occlusive disease of the internal carotid artery. *J Nucl Med* 1988;29:311–319.
15. Herold S, Brown MM, Frackowiak RSJ, Mansfield AO, Thomas DJ, Marshall J. Assessment of cerebral haemodynamic reserve: correlation between PET and CO_2 reactivity measured by the intravenous ^{133}xenon injection technique. *J Neurol Neurosurg Psychiatry* 1988;51:1045–1050.
16. Reich T, Rusinek H. Cerebral cortical and white matter reactivity to carbon dioxide. *Stroke* 1989;20:453–457.
17. Kuhl DE, Barrio JR, Huang SC, et al. Quantitative local cerebral blood flow by N-isopropyl-p-[^{123}I] iodoamphetamine (IMP) tomography. *J Nucl Med* 1982;23:196–203.
18. Kanno I, Uemura K, Higano S, et al. Oxygen extraction fraction at maximally vasodilated tissue in the ischemic brain estimated from the regional CO_2 responsiveness measured by positron emission tomography. *J Cereb Blood Flow Metab* 1988;8:227–235.
19. Leblanc R, Tyler JL, Mohr G, et al. Hemodynamic and metabolic effects of cerebral revascularization. *J Neurosurg* 1987;66:529–535.
20. Yamauchi H, Fukuyama H, Kimura J, Konishi J, Kameyama M. Hemodynamics in internal carotid artery occlusion examined by positron emission tomography. *Stroke* 1990;21:1400–1406.
21. Ringelstein EB, Sievers C, Ecker S, Schneider PA, Otis SM. Noninvasive assessment of CO_2-induced cerebral vasomotor response in normal individuals and patients with internal carotid artery occlusions. *Stroke* 1988;19: 963–969.
22. Lassen NA, Andersen AR, Friberg L, Paulson OB. The retention of [^{99m}Tc]-d,l-HM-PAO in the human brain after intracarotid bolus injection: a kinetic analysis. *J Cereb Blood Flow Metab* 1988;8:S13–22.
23. Ballinger JR, Gulenchyn KY, Reid RH. Radiopharmaceutical factors in variable quality of ^{99m}Tc-HMPAO images of the brain. *J Nucl Med* 1990; 31:118–122.
24. Choksey MS, Costa DC, Iannotti F, Ell PJ, Crockard HA. Technetium-99m-HMPAO SPECT and cerebral blood flow: a study of CO_2 reactivity. *Nucl Med Commun* 1989;10:609–618.
25. Buell U, Schicha H. Nuclear medicine to image applied pathophysiology: evaluation of reserves by emission computerized tomography. *Eur J Nucl Med* 1990;16:129–135.
26. Sullivan HG, Kingsbury TB, Morgan ME, et al. The rCBF response to diamox in normal subjects and cerebrovascular disease patients. *J Neurosurg* 1987;67:525–534.
27. Gitelman DR, Prohovnik I, Tatemichi TK. Safety of hypercapnic challenge: cardiovascular and neurologic considerations. *J Cereb Blood Flow Metab* 1991;11:1036–1040.
28. Ringelstein EB, Eyck SV, Mertens I. Evaluation of cerebral vasomotor reactivity by various vasodilating stimuli: comparison of CO_2 to acetazolamide. *J Cereb Blood Flow Metab* 1992;12:162–168.
29. Knop J, Thie A, Fuchs C, Siepmann G, Zeumer H. Technetium-99m-HMPAO-SPECT with acetazolamide challenge to detect hemodynamic compromise in occlusive cerebrovascular disease. *Stroke* 1992;23:1733–1742.
30. European Carotid Surgery Trialists' Collaborative Group. MRC european carotid surgery trial: interim results for symptomatic patients with severe (70%–99%) or with mild (0%–29%) carotid stenosis. *Lancet* 1991;337:1235–43.
31. North American Symptomatic Carotid Endarterectomy Trial Collaborators. Beneficial effect of carotid endarterectomy in symptomatic patients with high grade carotid stenosis. *N Engl J Med* 1991;325:445–53.
32. Matsuda H, Higashi S, Kinuya K, et al. SPECT evaluation of brain perfusion reserve by the acetazolamide test using Tc-99m HMPAO. *Clin Nucl Med* 1991;16:572–579.
33. Hashikawa K, Itagaki Y, Matsumoto M, et al. The effect of hypercapnia and hyperventilation on cerebral circulation in the patients with moyamoya disease [Abstract]. *J Nucl Med* 1993;34(suppl 5):63P.