and neglect after subcortical stroke: neuropsychological and cerebral perfusion study. *J Neurol Neurosurg Psychiatry* 1988;51:1269–1276.
27. Waldemar G, Hasselbalch SG, Andersen AR, et al. Technetium-99m-d,l-HMPAO and SPECT of the brain in normal aging. *J Cereb Blood Flow Metab* 1991;11:508–521.
28. Mountz JM. A method of analysis of SPECT blood flow image data for comparison with computed tomography. *Clin Nucl Med* 1989;14:192–196.
29. Davis SM, Chua MG, Lichtenstein M, Rossiter SC, Binns D, Hopper JL. Cerebral hypoperfusion in stroke prognosis and brain recovery. *Stroke* 1993;24:1691–1696.
30. Hoffman EJ, Cutler PD, Kigby WM, Mazziotta JC. Three-dimensional phantom to simulate cerebral blood flow and metabolic images for PET. *IEEE Trans Nucl Sci* 1990;37:616–620.
31. Hoffman EJ, Cutler PD, Guerrero TM, Digby WM, Mazziotta JC. Assessment of accuracy of PET utilizing a three-dimensional phantom to simulate the activity distribution of [^{18}F]fluorodeoxyglucose uptake in the human brain. *J Cereb Blood Flow Metab* 1991;11:A17–A25.
32. Siemens Microdelta User Manual, Revision 1. 1992;13–18.
33. Infeld B, Davis SM, Lichtenstein M, Mitchell PJ, Hopper JL. Crossed cerebellar diaschisis and brain recovery after stroke. *Stroke* 1995;26:90–95.
34. Blokland KA, Reiber HH, Pauwels EK. Quantitative analysis in single-photon emission computed tomography. *Eur J Nucl Med* 1992;19:47–61.
35. Tsui BM, Zhao X, Frey EC, McCartney WH. Quantitative single-photon emission computed tomography: basics and clinical considerations. *Semin Nucl Med* 1994;24: 38–65.
36. King MA, Long DT, Brill AB. SPECT volume quantitation: influence of spatial resolution, source size, shape and voxel size. *Med Phys* 1991;18:1016–1024.
37. Hoffman EJ, Huang SC, Phelps ME. Quantitation in positron emission computed tomography 1. Effect of object size. *J Comput Assist Tomogr* 1979;3:299–308.
38. Williams PL, Warwick R, eds. *Gray's anatomy*, 36th ed. Edinburgh: Churchill Livingstone; 1980:1002–1009.
39. Lenzi GL, Jones T, McKenzie CG, Buckingham PD, Clark JC, Moss S. Study of regional cerebral metabolism and blood flow relationships in man using the method of continuously inhaling oxygen-15 and oxygen-15 labeled carbon dioxide. *J Neurol Neurosurg Psychiatry* 1978;41:1–10.
40. Lenzi GL, Frackowiak RS, Jones T, et al. $CMRO_2$ and CBF by the oxygen-15 inhalation technique. Results in normal volunteers and cerebrovascular patients. *Eur Neurol* 1981;20:285–290.
41. Frackowiak RS, Lenzi GL, Jones T, Heather JD. Quantitative measurement of regional cerebral blood flow and oxygen metabolism in man using ^{15}O and positron emission tomography: theory, procedure and normal values. *J Comput Assist Tomogr* 1980;4: 727–736.
42. Baron JC, Bousser MG, Comar D, Soussaline F, Castaigne P. Noninvasive tomographic study of cerebral blood flow and oxygen metabolism in vivo. Potentials, limitations and clinical applications in cerebral ischemic disorders. *Eur Neurol* 1981;20:273–284.
43. Devous MD Sr, Stokely EM, Chehabi HH, Bonte FJ. Normal distribution of regional cerebral blood flow measured by dynamic single-photon emission tomography. *J Cereb Blood Flow Metab* 1986;6:95–104.
44. Davis SM, Chua MG, Lichtenstein M, Rossiter SC, Tress BM, Hopper JL. Perfusion changes with recovery after stroke [Abstract]. *Aust N Z J Med* 1993;23:568.
45. Podreka I, Baumgartner C, Suess E, et al. Quantification of regional cerebral blood flow with IMP-SPECT. Reproducibility and clinical relevance of flow values. *Stroke* 1989;20:183–191.
46. Isaka Y, Itoi Y, Imaizumi M, Ashida K, Okamoto M, Iiji O. Quantitation of rCBF by ^{99m}Tc-hexamethylpropylene amine oxime single-photon emission computed tomography combined with ^{133}Xe CBF. *J Cereb Blood Flow Metab* 1994;14:353–357.

Test/Retest Reproducibility of Iodine-123-βCIT SPECT Brain Measurement of Dopamine Transporters in Parkinson's Patients

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Iodine-123-β-CIT has been used as a probe of dopamine transporters in Parkinson's disease patients using SPECT. We studied the test/retest reproducibility of SPECT measures in Parkinson's disease patients and healthy controls obtained after injection of [^{123}I]β-CIT in part to assess the utility of this tracer for longitudinal evaluation of striatal dopamine transporters as a marker of disease progression. **Methods:** Seven Parkinson's disease patients and seven healthy control subjects participated in two [^{123}I]β-CIT SPECT scans separated by 7–21 days. Subjects were imaged at 24 hr post injection of 360 MBq (9.7 mCi) of [^{123}I]β-CIT. Two outcome measures were evaluated; 1) the ratio of specific striatal (activity associated with DA transporter binding) to nondisplaceable uptake, also designated V3," and 2) the total specific striatal uptake (%SSU) expressed as a percentage of injected radiotracer dose. For both measures, test/retest variability was calculated as the absolute difference of test minus retest divided by the mean of test/retest and expressed as a percent. In addition, the reproducibility of left and right striatal asymmetry and putamen:caudate ratios were determined. **Results:** The two outcome measures demonstrated excellent test/retest reproducibility for both the Parkinson's disease and healthy subject groups with variability of striatal V3" = 16.8 ± 13.3% and percent striatal uptake = 6.8 ± 3.4% for Parkinson's disease patients and V3" = 12.8 ± 8.9% and %SSU = 7.0 ± 3.9% for control subjects. There were no statistically significant differences in test/retest variability between control subjects and Parkinson's disease patients for either outcome measure. The reproducibility of left/right asymmetry indices and putamen-to-caudate ratios showed no patient versus control subject differences. The asymmetry index had greater test/retest variability than the other outcome measures. **Conclusion:** These data suggest that SPECT imaging performed at 24 hr postinjection of [^{123}I]β-CIT permits calculation of reliable and reproducible measures of dopamine transporters in both Parkinson's disease patients and control subjects and supports the feasibility of using [^{123}I]β-CIT in the evaluation of disease progression in Parkinson's disease.

Key Words: iodine-123; SPECT; dopamine transporters; Parkinson's disease

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Iodine-123-β-CIT ([^{123}I] 2β-carbomethoxy-3β-(4-iodophenyl-tropane) binds with high affinity to dopamine (DA) (IC50 = 1.6 n*M*) and serotonin (IC50 = 3.78 n*M*) transporters and has been used as a SPECT probe in human and nonhuman primates (*1–6*). In baboons, striatal activity is associated with binding to the DA transporters based on dynamic SPECT studies demonstrating displacement of this activity after administration of dopamine transporter-selective, but not serotonin transporter-selective agents (*3*). After intravenous administration of [^{123}I]β-CIT in humans, decay-corrected striatal time-activity data showed a prolonged time to highest uptake occurring by 12–18 hr post-tracer injection and very slow rates of striatal washout. Occipital and free parent plasma time-activity data reached a plateau earlier than striatum and also demonstrated extremely

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TABLE 1
Subject Demographics

Subject no.	Age (yr)	Gender	Hoehn-Yahr Stage	UPDRS		L-Dopa/carbidopa		DA agonist			Selegiline
				Total	Motor	Sinemet	Sinemet-CR	Amantadine	Pergolide	Bromocriptine	
PD-1	57	M	1	19	14						
PD-2	72	M	2	43	26.5		800		4		10
PD-3	78	F	4	58	34	450	300			10	
PD-4	67	M	1.5	24	11			200			
PD-5	63	M	2	29	17	200	300		3		10
PD-6	66	M	2	41	23	300	600				5
PD-7	57	M	3	41.5	26	450		200	7.5		
HS-1	24	F									
HS-2	39	M									
HS-3	19	F									
HS-4	37	M									
HS-5	34	M									
HS-6	62	M									
HS-7	74	F									

PD = Parkinson's disease subject; HS = healthy subject.

slow rates of washout. The protracted steady levels of parent activity in plasma and activity within brain compartments closely approximated equilibrium binding conditions at the DA transporter (*5*). Under such conditions, a simple ratio of specific striatal-to-background activity calculated during the plateau phase of uptake provides an outcome measure that is proportional to dopamine transporter density (*5*).

Using this outcome measure, healthy subjects show expected age-related reduction in uptake as a function of normal aging (*7*). Idiopathic Parkinson's disease patients demonstrated reduced [^{123}I]β-CIT uptake compared with age and gender-matched healthy control subjects (*2,8,9*). In addition, patients with Hoehn-Yahr Stage I disease (hemiparkinsonism) showed reductions of uptake in the striatum contralateral to symptom side and ipsilateral as well suggesting the test is sensitive to changes in DA transporters before the appearance of clinical symptoms (*10*). In a series of 28 Parkinson's disease subjects, [^{123}I]β-CIT SPECT uptake was correlated with symptom severity as measured by the United Parkinson's Disease Rating Scale (UPDRS) (*8*).

To assess the applicability of [^{123}I]β-CIT SPECT imaging as a potential marker in the evaluation of disease progression in Parkinson's disease patients, we undertook an initial investigation of the test/retest reproducibility of SPECT outcome measures in healthy control subjects (*11*). This study demonstrated robust test/retest reproducibility of the two outcome measures measured at 18–24 hr post-tracer injection: the ratio of specific to nondisplaceable uptake as well as the specific striatal uptake expressed as a percentage of the injected dose. In the present evaluation, we undertook to extend this study to Parkinson's disease patients to further assess the suitability of [^{123}I]β-CIT SPECT for serial evaluation of Parkinson's disease patients.

MATERIALS AND METHODS

Radiopharmaceuticals

High specific activity [^{123}I]β-CIT was prepared from the corresponding trimethylstannyl precursor (*1*) and high radionuclidic purity [^{123}I]NaI as described previously (*12*). Radiochemical purity was greater than 94% as measured with high-performance liquid chromatography (HPLC), and the specific activity was >5,000 Ci/mmol.

Subjects

Seven patients (age 65.7 ± 7.6 yr, with these and subsequent measured expressed as mean ± s.d.) with idiopathic Parkinson's disease (Hoehn-Yahr Stages 1–3) (Table 1) were enrolled in the study after giving informed consent. Inclusion criteria included age greater than 35 yr and at least two of the following: bradykinesia, resting tremor, rigidity, postural instability, or freezing phenomenon (one of which is rest tremor or bradykinesia). All patients were evaluated in the Yale-New Haven Hospital General Clinical Research Center using the modified Core Assessment Program for Intracerebral Transplant (CAPIT) protocol (*13*). Motor function was rated using the UPDRS (*14*) and four timed motor tasks after 12-hr medication withdrawal and 1 hr following oral administration of carbidopa/L-dopa (25/100 mg). Response to L-dopa challenge was defined as a 33% improvement in UPDRS score or in one timed motor test. Patients who failed to respond were rechallenged with a repeat single oral dose carbidopa/L-dopa (25/250 mg) and reexamined 1 hr later using these criteria. Those patients showing no response to L-dopa were excluded from the study. Subjects received their usual antiparkinson medication after the evaluation and during the SPECT study. Subjects were excluded if taking medications known to have a significant effect on the dopamine transporter (e.g., benztropine). No changes in medications were permitted over the duration of the study. Subjects received oral administration of supersaturated potassium iodide (SSKI, 800 mg) before injection of 358 ± 19 MBq (9.7 ± 0.5 mCi) [^{123}I]β-CIT. Each subject participated in two SPECT studies separated by 7–21 days.

Seven healthy subjects previously studied and reported (*11*) served as control subjects. Healthy subjects underwent same image acquisition protocol on the same SPECT camera.

Data Acquisition and Analysis

Four fiducial markers each filled with 1–4 μCi [^{99m}Tc]NaTcO4 were attached to both sides of the subject's head at the level of the canthomeatal line before imaging to facilitate posthoc reorientation of transaxial images. At 24 hr postinjection of [^{123}I]β-CIT, 120 raw projection images were acquired into a 64 × 64 matrix. Three 15-min acquisitions were obtained 24 hr postinjection.

Raw data were reconstructed from photopeak counts within a 20% symmetric energy window centered around 159 keV using a Butterworth filter (power factor = 10, cutoff = 1 cm). Transaxial images were reoriented parallel to the canthomeatal plane and

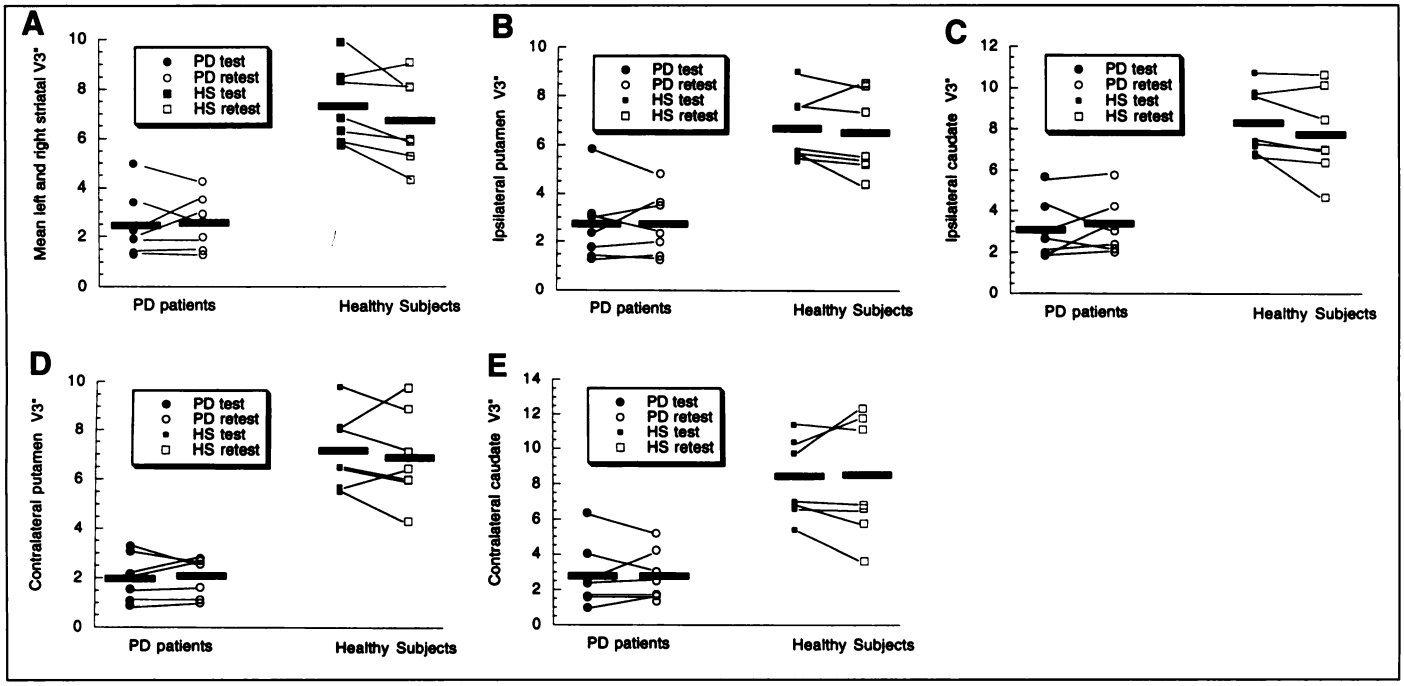

FIGURE 1. Test/retest measures of V3" in seven Parkinson's disease patients and seven healthy control subjects. (A) Overall striatal ratio with individual subject test/retest scores indicated by symbols joined by a line. Panels B–E are scores for ipsilateral (right in control subjects) and contralateral (left in control subjects) caudate and putamen as indicated. Means are indicated by the dark bars.

attenuation-corrected using Chang zero order correction (*15*) based on an ellipse fit to brain using a linear attenuation factor (μ = 0.15 cm^{-1}) determined empirically from an ^{123}I-containing distributed source phantom.

Two outcome measures were evaluated as previously described (*11*): the ratio of specific striatal uptake to nondisplaceable uptake (designated V3") and the total left and right specific striatal uptake (designated %SSU) expressed as a percent of injected radioactivity. A trained research technologist blind to the clinical status of the subject performed the image processing. For V3," four contiguous transaxial slices representing the most intense striatal uptake were summed. A standard region of interest (ROI) template was constructed based on coregistered MRI scans obtained from previous [^{123}I]β-CIT studies in four healthy control subjects. This template included regions for left and right striatum, frontal cortex, and occipital cortex. Small variations in individuals' brain required movement of the ROI's within the template without changing the individual ROI shape or pixel size. Data were expressed as counts/pixel/minute for each brain region. Estimates of specific striatal uptake were made by subtracting occipital counts/pixel/minute from total striatal counts/pixel/minute based on the low density of monoamine transporters in the occipital cortex. V3," the ratio of striatal activity specifically bound to transporters divided by nondisplaceable activity is equal to the binding potential (BP) divided by the nonspecifically-bound compartment distribution volume (V2) under conditions of equilibrium binding; i.e., when the concentration of parent compound is unchanging in plasma, receptor-bound and nonspecifically-bound brain compartments. This method assumes equivalence of nondisplaceable uptake in striatum and occipital cortex. V3" was derived by dividing the operationally-defined specific striatal uptake by occipital uptake, this measure is equal to total striatal activity-to-occipital activity minus 1. The final ratio was calculated as the mean of all V3" measurements over 24 hr (three scans total)) for left and right caudate and putamen. Based on these mean V3" ratios, two derived measures were calculated: the ratio of putamen-to-caudate for both left and right striata and an asymmetry index (AI) defined as the absolute value of the test minus retest condition divided by the mean of the test and retest conditions and expressed as a percent.

The percent uptake in striatal and occipital ROIs used a larger number of transaxial slices (n = 14) than the ratio of specific to nondisplaceable uptake (n = 4) in order to recover all specific activity associated with striatum. In all subjects the summed slices included one or two slices extending beyond visually-identified striatum. A standardized, elliptical ROI slightly larger than the visually identified striatum was placed over the summed slices over regions corresponding to left and right striatum and occipital cortices bilaterally. The size of these ROIs (left striatum, right striatum, left occipital, right occipital) were identical and, therefore, produced identical volumes for measurement of total counts in striatal regions and occipital cortex. The same template was used for all subjects. As in the previous analyses, nondisplaceable striatal uptake was estimated from occipital counts. Total counts within the occipital volume of interest were subtracted from the total counts within the striatal volumes to generate a measure of counts associated with specific striatal uptake. In this instance the measure is more akin to total receptor number rather than receptor concentration provided by V3". Counts were corrected for physical decay and converted to μCi of activity based on ^{123}I distributed source cylindrical phantoms (length = 12.9 cm, width = 10.5 cm, total volume = 1000 ml) containing ~500 μCi of activity. The mean of the three percent specific striatal uptake measurements made at 24 hr post-tracer injection are reported.

Statistical Analyses

Repeated measures ANOVA was used for evaluating within and between group differences for V3" obtained in left and right caudate and putamen, %SSU and percent occipital uptake determined from the mean of the three scans at the 24 hr time point. Post hoc analyses used two tailed Dunnett's Student's t-test with significance at the $p < 0.05$ level. For both outcome measures V3" and %SSU, within-subject variability between test and retest conditions was calculated as the absolute value of the difference of the test and retest measure divided by the mean of test and retest

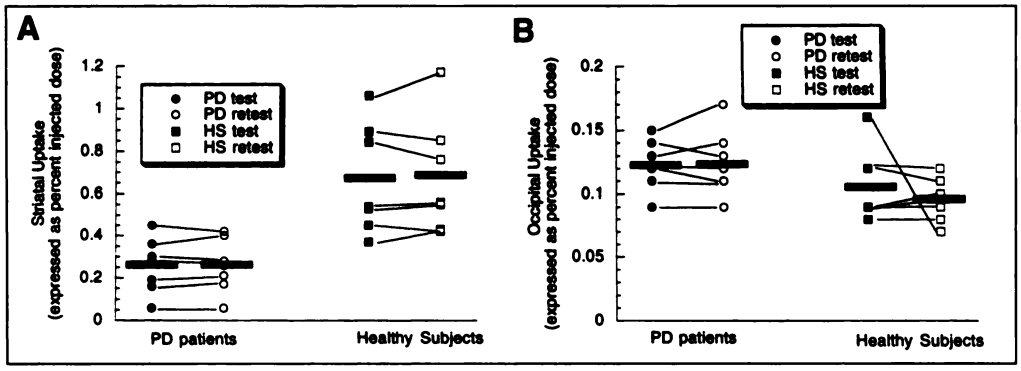

FIGURE 2. Test/retest measures of percent specific striatal uptake (%SSU) and percent occipital uptake in seven Parkinson's disease patients and seven healthy control subjects.

and expressed as a percentage. ANOVA using one within and one between group factor was calculated for the Parkinson's disease and control group using the variability data for V3" calculated for each striatal region, %SSU and percent occipital uptake as well as the variability measures for the two measures derived from V3" (putamen:caudate ratio and asymmetry indices). For these analyses in Parkinson's disease patients, the designation of contralateral striatum was made for the side opposite initial symptom presentation. For the healthy control subjects, left striatal regions were arbitrarily compared with the contralateral patient side.

RESULTS

Figure 1A indicates the overall mean striatal V3" test/retest values for Parkinson's disease and control subjects. For the Parkinson's disease group, the mean V3" of the test condition was 2.5 ± 1.3 and retest 2.6 ± 1.1. For the healthy group, the mean of the V3" test was 7.3 ± 1.6 and retest 6.6 ± 1.8. Repeated measures ANOVA showed significant between group ($F = 35.24$ $p = 0.0001$), but not within-subject ($F = 2.47$ $p = 0.14$) differences, which was expected because of the lower striatal activity in the Parkinson's disease group. Posthoc analysis using a two-tailed Dunnet's Student's t-test was significant for the between group effect.

Test/retest V3" for ipsilateral and contralateral caudate and putamen are also indicated in Figure 1B–E. The mean test/retest for Parkinson's disease subjects were 2.95 ± 1.42 and 3.17 ± 1.35 (test and retest conditions, respectively) in ipsilateral caudate, 2.89 ± 1.86 and 2.90 ± 1.45 in contralateral caudate, 2.60 ± 1.57 and 2.55 ± 1.31 in ipsilateral putamen, and 2.12 ± 0.92 and 2.24 ± 0.80 in contralateral putamen. For the healthy subjects, the mean test/retest V3" were 8.33 ± 1.66 and 7.77 ± 2.12 (test and retest conditions, respectively) in right caudate, 8.18 ± 2.27 and 8.30 ± 3.41 in left caudate, 6.61 ± 1.44 and 6.39 ± 1.69 in right putamen, and 7.16 ± 1.55 and 6.91 ± 1.85 in left putamen. Repeated measures ANOVA showed significant between group but not within-subject differences for ipsilateral (right) caudate ($F = 32.47$ $p = 0.0001$, $F = 0.50$ $p = 0.49$, between and within subject effects, respectively), contralateral (left) caudate ($F = 19.91$ $p = 0.0008$, $F = 0.04$ $p = 0.85$), ipsilateral (right) putamen ($F = 23.55$ $p = 0.0004$, $F = 0.30$ $p = 0.59$), and contralateral (left) putamen ($F = 52.06$ $p = 0.0001$, $F = 0.20$ $p = 0.66$). Posthoc analysis using a two-tailed Dunnet's Student's t-test was significant for the between group effect in all four striatal regions.

The test/retest scores for Parkinson's disease patients and control subjects for %SSU and percent occipital uptake are

TABLE 2
Test/Retest Variability for Striatal Ratio and Percent Striatal Uptake

	Test/Retest Variability					
	Specific striatal:nondisplaceable uptake (V3")				Percent uptake	
Subject	ipsi caud	ipsi putamen	contra caud	contra putamen	%SSU	%OCCIPITAL
PD Patient 1	8.0	10.1	16.7	11.2	6.9	8.7
PD Patient 2	32.9	29.2	28.8	18.7	7.4	7.4
PD Patient 3	33.7	42.9	46.6	25.4	10.5	12.5
PD Patient 4	51.2	13.9	51.4	26.1	10.0	0.0
PD Patient 5	10.9	10.9	4.9	4.7	6.9	7.4
PD Patient 6	1.8	19.4	20.4	18.6	0.0	0.0
PD Patient 7	9.0	8.3	6.3	3.7	6.1	0.0
MEAN	**21.1**	**19.2**	**25.0**	**15.5**	**6.8**	**5.1**
%COV	**86.7**	**65.9**	**73.3**	**59.2**	**50.5**	**99.2**
Healthy Subject 1	4.1	1.5	3.1	12.2	9.9	78.3
Healthy Subject 2	3.8	12.4	12.7	17.9	5.5	10.5
Healthy Subject 3	12.2	3.4	23.8	12.2	10.0	8.7
Healthy Subject 4	37.3	24.3	39.7	24.1	1.8	10.5
Healthy Subject 5	2.4	3.8	5.5	8.7	4.6	0.0
Healthy Subject 6	1.0	6.7	2.4	9.7	4.5	0.0
Healthy Subject 7	15.0	4.1	15.4	7.9	12.7	0.0
MEAN	**10.8**	**8.0**	**14.7**	**13.2**	**7.0**	**15.4**
%COV	**118.1**	**99.8**	**91.6**	**44.2**	**55.3**	**182.5**

Variability is absolute value (test − retest)/(mean test and retest) expressed as a percent. Ipsilateral is arbitrarily designated as right in healthy controls. ipsi = ipsilateral; contra = contralateral; caud = caudat; and %SSU = percent specific striatal uptake.

TABLE 3
Test/Retest Variability for Putamen: Caudate Ratio and Striatal Asymmetry

	Test/Retest Variability			
	Ipsilateral (right) putamen-to-caudate ratio	Contralateral (left) putamen-to-caudate ratio	Ipsilateral (right) asymmetry index	Contralateral (left) asymmetry index
PD Patient 1	18.0	27.8	85.2	58.4
PD Patient 2	3.9	10.2	189.3	112.6
PD Patient 3	9.5	21.8	185.7	102.4
PD Patient 4	38.0	26.1	0.2	38.0
PD Patient 5	21.7	0.3	95.9	36.3
PD Patient 6	21.1	1.8	10.2	1.3
PD Patient 7	0.7	2.6	12.6	20.7
Mean	**16.1**	**12.9**	**82.7**	**52.8**
%COV	**78.8**	**93.1**	**97.6**	**78.2**
Healthy Subject 1	5.6	9.0	48.1	103.4
Healthy Subject 2	8.6	5.2	85.7	53.2
Healthy Subject 3	8.8	35.8	182.9	38.1
Healthy Subject 4	13.3	15.9	9.9	8.9
Healthy Subject 5	1.4	3.1	74.2	30.3
Healthy Subject 6	5.7	7.3	29.0	44.2
Healthy Subject 7	10.9	7.5	4.3	40.2
Mean	**7.8**	**12.0**	**62.0**	**45.5**
%COV	**50.5**	**93.6**	**99.1**	**63.9**

Ipsilateral is arbitrarily designated as right in healthy control subjects.

indicated in Figure 2. In the Parkinson's disease, group the mean %SSU was 0.257% ± 0.131% and 0.257% ± 0.126% for the test and retest conditions, respectively. For the healthy control subjects the mean %SSU was 0.669% ± 0.260% and 0.677% ± 0.270% for test and retest scans, respectively. Similar to V3," %SSU for the mean of three scans at 24 hr postinjection demonstrated statistically significant differences between groups ($F = 14.14$ $p = 0.0027$) but no within-subject differences for the test/retest conditions ($F = 0.11$ $p = 0.74$). Two-tailed Dunnett's test confirmed significant differences between the Parkinson's disease and control groups.

The mean percent occipital uptake was 0.122% ± 0.019% and 0.124% ± 0.026% for the Parkinson's disease test and retest conditions, respectively. Healthy subjects had a mean percent occipital uptake of 0.107% ± 0.027% and 0.096% ± 0.017% for test and retest conditions, respectively. Occipital percent uptake showed no significant within subject effect ($F = 0.51$, $p = 0.49$). The between group effect was significant ($F = 4.71$, $p = 0.05$), although Dunnett's test did not confirm significant differences between the healthy and Parkinson's disease groups.

Variability Scores

Table 2 summarizes the variability data for both striatal outcome measures and percent occipital uptake. The overall striatal V3" had a test/retest variability of 12.8 ± 9.0% for the mean of three scans at 24 hr in the healthy subjects and 16.8 ± 13.3% for the Parkinson's disease patients. ANOVA demonstrated no significant differences in the variability of V3" ($F = 0.43$, $p = 0.52$). Considering each striatal region separately, ANOVA showed no significant between subject effects for ipsilateral (right) caudate ($F = 1.48$ $p = 0.25$), contralateral (left) caudate ($F = 1.45$ $p = 0.25$), ipsilateral (right) putamen ($F = 3.92$ $p = 0.07$) and contralateral (left) putamen ($F = 0.30$ $p = 0.59$).

The test/retest variability scores for putamen-to-caudate ratios and asymmetry indices in healthy subjects and Parkinson's disease patients are indicated in Table 3. The asymmetry index demonstrated greater test/retest variability than putamen:caudate ratios or regional striatal V3". ANOVA performed on the variability data showed no significant between subject differences for ipsilateral (right) caudate putamen:caudate ratio ($F = 2.77$, $p = 0.12$), contralateral (left) putamen:caudate ratio ($F = 0.02$, $p = 0.88$), caudate asymmetry index ($F = 0.29$, $p = 0.60$) or putamen asymmetry index ($F = 0.15$, $p = 0.71$).

The test/retest variability of %SSU was 7.0 ± 3.8% and 6.8 ± 3.4% for the control subjects and Parkinson's disease patients, respectively. The test/retest variability in occipital uptake was 15.4 ± 28.1% and 5.1 ± 5.1% for the healthy subjects and Parkinson's disease patients, respectively. ANOVA showed no significant differences in %SSU ($F = 0.01$, $p = 0.93$), or percent occipital uptake ($F = 0.90$, $p = 0.36$) between Parkinson's disease and healthy subjects.

DISCUSSION

Similar to healthy subjects previously described, Parkinson's disease patients evidence good test/retest reproducibility for the two [^{123}I]β-CIT SPECT outcome measures evaluated; the ratio of specific to nondisplaceable uptake (V3") and %SSU. These measures potentially have different sources of test/retest error. While not achieving statistical significance in previous analyses (*11*), both healthy subjects showed lower test/retest variability for %SSU compared with V3". This might be attributable to the compounding of error in V3" which represents a ratio of measurements in striatum to occipital background, where the background region has very few counts and therefore is liable to greater measurement error. %SSU uses a measure of the background counts but subtracts these from total striatal counts, hence, there is less statistical propagation of error. Nonetheless, V3" would be the theoretically superior measure compared with %SSU given the potential confound of variability in peripheral clearance of tracer and the requirement of accurate calibration of the instrument for %SSU. In addition, if one assumes equivalence of nondisplaceable uptake in the striatum between

subjects, the measure V3" is directly related to the binding potential defined as Bmax/Kd (*5*).

The fact that percent occipital uptake was not significantly different between the two groups indirectly supports:

1. The assumption of the comparability of nondisplaceable tracer uptake between patients and control subjects.
2. The validity of comparing %SSU in healthy and Parkinson's disease subjects.

The comparability of percent occipital uptake indirectly suggests there were no large differences in peripheral radiotracer clearance between the groups. The reproducibility of percent occipital uptake further suggests that in these test/retest scans, measured within a short time period, there are no significant within-subject differences in clearance. Nonetheless, caution should be exercised when %SSU is used as an outcome measure in an elderly patient population on medications that could alter rates of radiopharmaceutical clearance from the vascular pool.

The variability of V3" measured for separate striatal regions (ipsilateral and contralateral caudate and putamen) shows less robust reproducibility than the mean V3" measures. Regional striatal measures are more relevant to studying Parkinson's disease than overall striatal measures given the greater loss of dopamine transporters in putamen compared with caudate in Parkinson's disease patients.

We hypothesized that Parkinson's disease patients would have poorer reproducibility of both V3" and %SSU compared to healthy control subjects because of lower striatal accumulation of tracer in the patients. However, the mean variability of V3," though higher in Parkinson's disease patients compared with control subjects, did not achieve statistical significance. There were no differences in the test/retest variability of %SSU or percent occipital uptake between the groups.

Reproducibility of imaging measures are dependent on the choice of SPECT instrument and parameters of scan acquisition and image processing. As previously demonstrated in healthy subjects (*11*), one source of error evident in our analysis is a ring artifact resulting from septal penetration by high energy photons of the collimator used in the study. This ring is in close proximity to the occipital background region and could contribute to error in occipital activity measures. The specific parameters of acquisition will also affect the reproducibility of the SPECT measures. In our previous study of healthy subjects we demonstrated slight improvement of V3" measures of variability when using scans obtained at 18 and 21 hr post injection in addition to the 24 hr measure. This study in Parkinson's disease patients used only scans obtained at 24 hr as this would most closely replicate the clinical conditions under which such a test would be performed. Issues of collimator sensitivity and amount of injected activity/scan duration will affect the reproducibility of quantitative outcome measures. The selection of a reconstruction filter and ROI size and placement will also affect the variability measures. Smoother filters and larger ROIs could result in better reproducibility, although at the cost of poor discrimination between caudate and putamen. One factor that can deleteriously influence the reproducibility of the putamen-to-caudate ratio is the application of large ROIs that may not have adequately separated counts from these regions. Finally, the healthy control subjects in this study were younger than the Parkinson's disease patients. Previous studies with [^{123}I]β-CIT SPECT reveal a decline in striatal uptake in healthy subjects as a function of normal aging (*7*). The significance of using a younger healthy cohort in the present investigation produces a smaller source of difference between the Parkinson's disease and healthy subjects than the underlying pathophysiological process. Both the effects of age and disease status would tend to increase the variability of the within-subject measures in the Parkinson's disease patients relative to control subjects. Nonetheless, no differences on the variability measures were demonstrated between the two groups.

Is the test/retest variability associated with [^{123}I]β-CIT measures in Parkinson's disease patients acceptable for serial studies of disease progression within an individual patient? Studies in Parkinson's disease patients suggest an annual change of 9–14 points in UPDRS points per year (*16,17*). Based on our earlier study of 28 Parkinson's disease patients across a range of disease severity (*8*), we performed a linear regression analysis on V3" from contralateral putamen plotted against total UPDRS score. We estimate a reduction in V3" of 1.2% for each point change in total UPDRS score. Hence, a 12-point change in total UPDRS would result in a reduction of striatal signal of 14%. It should be underscored that these are crude estimates based on cross-sectional data. Variability in the rate of disease progression between patients is high and dependent of the stage of illness. Within the individual, some evidence suggests that the rate of progression as measured by [^{18}F]F-dopa PET is faster in the early stage of illness (*18*), although this was not confirmed in a larger sample that included more later stage Parkinson's disease patients (personal communication). Studies designed to demonstrate the utility of [^{123}I]β-CIT SPECT as an objective marker of disease progression should carefully consider the stage of illness. Better estimates would use progression data obtained from serial [^{123}I]β-CIT SPECT scans acquired during different stages of illness, and such data is being acquired by our group.

CONCLUSION

Iodine-123-β-CIT SPECT imaging in Parkinson's disease patients and healthy subjects demonstrates good test/retest reproducibility for two outcome measures. This supports the feasibility of using [^{123}I]β-CIT for SPECT measurement of dopamine transporters in the evaluation of neuropsychiatric illness affecting dopamine neuronal function.

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REFERENCES

1. Neumeyer JL, Wang S, Milius RA, et al. Iodine-123-2-β-carbomethoxy-3-β-(4-iodophenyl)-tropane (β-CIT): high affinity SPECT radiotracer of monoamine reuptake sites in brain. *J Med Chem* 1991;34:3144–3146.
2. Innis R, Seibyl J, Scanley B, et al. SPECT imaging demonstrates loss of striatal monoamine transporters in Parkinson's disease. *Proc Natl Acad Sci* 1993;90:11965–11969.
3. Laruelle M, Baldwin R, Malison R, et al. SPECT imaging of dopamine and serotonin transporters with [^{123}I]β-CIT: pharmacological characterization of brain uptake in nonhuman primates. *Synapse* 1993;13:295–309.
4. Seibyl J, Wallace E, Smith E, et al. Whole-body biodistribution, radiation absorbed dose and brain SPECT imaging with [^{123}I]β-CIT in healthy human subjects. *J Nucl Med* 1994;35:764–770.
5. Laruelle M, Wallace E, Seibyl J, et al. Graphical, kinetic and equilibrium analyses of in vivo [^{123}I]β-CIT binding to dopamine transporters in healthy human subjects. *J Cereb Blood Flow Metab* 1994;14:982–994.
6. Brücke T, Kornhuber J, Angelberger P, Asenbaum S, Frassine H, Podreka I. SPECT imaging of dopamine and serotonin transporters with [^{123}I]beta-CIT: binding kinetics in the human brain. *J Neural Trans* 1993; 94:137–46.
7. van Dyck C, Seibyl J, Malison R, et al. Age-related decline in striatal dopamine transporter binding with iodine-123-β-CIT SPECT. *J Nucl Med* 1995;36:1175–1181.

8. Seibyl J, Marek K, Quinlan D, et al. Decreased SPECT [^{123}I]β-CIT striatal uptake correlates with symptom severity in Parkinson's disease. *Ann Neurol* 1995;38:589–598.
9. Rinne J, Kuikka J, Bergstrom K, Rinne U. Striatal dopamine transporter in different disability stages of Parkinson's disease studied with [^{123}I]β-CIT SPECT. *Parkinsonism Relat Dis* 1995;1:47–51.
10. Marek K Seibyl J, Zoghbi S, et al. Iodine-123-β-CIT SPECT imaging demonstrates bilateral loss of dopamine transporters in hemi-parkinson's disease. *Neurology* 1996;46:231–237.
11. Seibyl J, Laruelle M, van Dyck C, et al. Reproducibility of [^{123}I]β-CIT SPECT brain measurement of dopamine transporters in healthy human subjects. *J Nucl Med* 1996;37:222–228.
12. Baldwin R, Zea-Ponce Y, Zoghbi S, et al. Evaluation of the monoamine uptake site ligand [^{123}I]methyl 3β-(4-iodophenyl)-tropane-2β-carboxylate (^{123}I)β-CIT in non-human primates: pharmacokinetics, biodistribution and SPECT brain imaging coregistered with MRI. *Nucl Med Biol* 1993;20:597–606.
13. Langston J, Widner H, Goetz CG, et al. Core assessment program for intracerebral transplantations. *Mov Dis* 1992;7:2–13.
14. Fahn S, Elton R, Members of the UPDRS Development Committee Unified Parkinson's disease rating scale. In: Fahn S, Marsen CD, Calne DB, Goldstein M, eds. *Recent developments in Parkinson's disease*, vol. 2. Florham Park, NJ: Macmillan Healthcare Information; 1987:153–164.
15. Chang LT. A method for attenuation correction in computed tomography. *IEEE Trans Nucl Sci* 1987;NS-25:638–643.
16. LeWitt PA. Clinical trials of neuroprotection in Parkinson's disease: long-term selegiline and alpha-tocopherol treatment. *J Neural Trans Suppl* 1994;43:171–181.
17. Lee C, Schulzer M, Mak E, et al. Clinical observations on the rate of progression of idiopathic parkinsonism. *Brain* 1994;117:501–507.
18. Morrish P, Sawle G, Brooks D. An [^{18}F]dopa-PET and clinical study of the rate of progression in Parkinson's disease. *Brain* 1996:in press.
19. Vingerhoets F, Snow B, Lee C, Schulzer M, Mak E, Calne D. Longitudinal fluorodopa positron emission tomographic studies of the evolution of idiopathic parkinsonism. *Ann Neurol* 1994;36:759–764.

Regional Methionine and Glucose Uptake in High-Grade Gliomas: A Comparative Study on PET-Guided Stereotactic Biopsy

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Gliomas are regionally heterogeneous tumors. The local relationship between histologic features and radiotracer uptake evaluated by PET should therefore influence analysis and interpretation of PET results on gliomas. This study explored this local relationship as a result of PET guidance of stereotactic biopsies. **Methods:** Local histology was confronted to the regional uptake of ^{18}F-2-fluoro-2-deoxy-D-glucose (^{18}F-FDG) and ^{11}C-methionine (^{11}C-MET) in 14 patients with high-grade glioma diagnosed during a procedure of PET-guided stereotactic biopsies. We analyzed the uptake of both tracers in regions of interest centered on the stereotactic coordinates of 93 biopsy samples. **Results:** A semiquantitative analysis revealed a significant regional correlation between ^{11}C-MET and ^{18}F-FDG uptakes. Uptake of both tracers was significantly higher on the site of tumor samples showing anaplastic changes than in the rest of the tumor. Presence of necrosis in anaplastic areas of the tumor significantly reduced the uptake of ^{11}C-MET. **Conclusion:** PET with ^{11}C-MET and ^{18}F-FDG may help to evaluate, in vivo, the metabolic heterogeneity of human gliomas. Anaplasia is a factor of increased uptake of both tracers, but microscopic necrosis in anaplastic areas influences their uptake differently. This finding probably relates to the differences in tracer uptake by non-neoplastic components of necrotic tumors. These results underline the complementary role of ^{18}F-FDG and ^{11}C-MET for the study of brain tumors and favors their use for stereotactic PET guidance of diagnostic or therapeutic procedures.

Key Words: glioma; PET; fluorine-18-fluorodeoxyglucose; methionine; stereotaxy

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Different radiotracers have been used with PET to help in the management of patients with glioma (*1,2*). These tracers may be classified into three groups: the markers of energetic metabolic pathways, the markers of protein and nucleic acid synthetic pathways and the radioligands for receptor imaging. Most investigators use tracers from the two first classes, e.g., ^{18}F-2-fluoro-2-deoxy-D-glucose (^{18}F-FDG) which assays glucose metabolism, and ^{11}C-methionine (^{11}C-MET) or other amino acid tracers, which assay amino acid transport and metabolism (*3–7*). Tracer choice depends on the goals pursued which may involve diagnosis, lesion delineation, grade and prognosis estimation and evaluation or prediction of response to treatment. For instance, it has been claimed that, for the definition of tumor limits, PET with ^{11}C-MET (MET-PET) is better than PET with ^{18}F-FDG (FDG-PET) or other modalities such as CT and MRI (*8,9*). To compare the characteristics of different PET tracers, it is essential to ascertain that the brain regions investigated during different PET procedures are similar, a condition which may be fulfilled using matching (*10*) or preferably stereotactic methods (*8,11–13*). Another important requisite for these comparative analyses is the definition, on the brain images, of regions of interest (ROIs) which adequately sample the tumor tissue. Since areas of high tracer uptake may be different with diverse tracers and since other imaging modalities, such as MRI, cannot ensure where the limits of the tumor are (*9*), histologic control of the regions concerned by the analysis seem to offer the best guarantee that this requirement is achieved. Furthermore, this histologic control allows confrontation of the multiple metabolic data with pathological features of the tumor. Therefore, we decided to apply a procedure of PET-guided stereotactic biopsies (*11*) to the comparison and histologic confrontation of PET information provided by two major tumor tracers used for the management of brain tumors: ^{18}F-FDG and ^{11}C-MET.

MATERIALS AND METHODS

Patients

A consecutive series of 19 patients suspected of having a brain tumor gave informed consent to undergo stereotactic biopsies guided by CT and by PET with successive injection of ^{11}C-MET and ^{18}F-FDG after a procedure which allows image data acquisi-

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