Decreased Striatal Monoaminergic Terminals in Multiple System Atrophy Detected with Positron Emission Tomography

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We examined the density of striatal presynaptic monoaminergic terminals, using a ligand for the type 2 vesicular monoamine transporter, $(+)$-$[^{11}C]$dihydrotetrabenazine, with positron emission tomography in 7 normal control subjects, 8 multiple system atrophy (MSA) patients with predominantly parkinsonian features (MSA-P), 8 MSA patients with principally cerebellar dysfunction (MSA-C), and 6 sporadic olivopontocerebellar atrophy (sOPCA) patients. The findings were correlated with the results of neurological evaluations and magnetic resonance imaging studies. Specific binding was significantly reduced in the putamen of all patient groups in the order MSA-P < MSA-C < sOPCA, compared with controls. Mean blood-to-brain ligand transport (K_1) was significantly decreased in the putamen of all patient groups and in the cerebellar hemispheres of MSA-C and sOPCA but not MSA-P groups, compared with controls. Significant negative correlations were found between striatal binding and the intensity of parkinsonian features and between cerebellar K_1 and the intensity of cerebellar dysfunction. The results suggest fundamental differences between MSA-P and MSA-C groups reflecting differential severity of degeneration of nigrostriatal and cerebellar systems in these two forms of MSA. The findings also show that some sOPCA patients have subclinical nigrostriatal dysfunction and are at risk of developing MSA with disease progression.

Multiple system atrophy (MSA) is a sporadic neurodegenerative disease of undetermined cause characterized by combinations of the parkinsonian symptoms of bradykinesia and rigidity that are poorly responsive or unresponsive to levodopa (ie, cerebellar ataxia, and autonomic insufficiency, frequently accompanied by signs of corticospinal tract disease).[1–10] Some MSA patients initially develop a parkinsonian syndrome (MSA-P) followed by autonomic dysfunction or cerebellar manifestations or both. In others, MSA begins with cerebellar manifestations (MSA-C) followed by autonomic dysfunction or parkinsonian features or both. MSA is a distinct entity clinically, neuropathologically, and biochemically.[1–4,11–20] The neuropathological findings consist principally of neuronal loss and gliosis in the caudate nucleus, putamen, external pallidum, substantia nigra, locus ceruleus, inferior olives, pontine nuclei, cerebellar Purkinje cells, and intermediolateral cell columns of the spinal cord.[21–25] Other structures showing variable involvement include the thalamus, vestibular

nucleus, dorsal vagal nucleus, corticospinal tracts, and anterior horn cells.[4,21–25] Oligodendroglial[26–30] and neuronal[31–34] intracytoplasmic and intranuclear inclusions are characteristic of MSA and are widely distributed in the central nervous system.

Sporadic olivopontocerebellar atrophy (sOPCA), also termed idiopathic sporadic cerebellar ataxia or idiopathic late-onset cerebellar ataxia, is a neurodegenerative disease of undetermined cause.[35–41] The symptoms consist of progressive ataxia of gait, speech, and limb movements accompanied by disturbances of extraocular movements and, in some patients, signs of corticospinal tract disease.[35–39] The neuropathological changes usually consist of neuronal loss and gliosis in the brainstem (inferior olives, pontine nuclei, and vestibular nuclei) and cerebellum (molecular, Purkinje cell, and granular layers).[22,40–42] Some patients who initially present with sOPCA later develop autonomic failure and parkinsonian features, indicating progres-

Table 1. Clinical Features in Patients with Multiple System Atrophy Presenting with Parkinsonian Features (MSA-P), Cerebellar Disorders (MSA-C), and Sporadic Olivopontocerebellar Atrophy (sOPCA)

Patient Diagnosis	1 MSA-P	2 MSA-P	3 MSA-P	4 MSA-P	5 MSA-P	6 MSA-P	7 MSA-P	8 MSA-P	9 MSA-C	10 MSA-C	11 MSA-C
Sex	M	F	M	M	M	M	F	M	F	M	F
Age (yr)	44	67	84	67	81	66	56	63	64	59	54
Duration of symptoms (yr)	1.5	3	4	2.5	5	2	7	4	6	5	7
Akinesia	++	+	++	++	+	+	+++	+++	+	+	+
Rigidity	++	+	+++	+	+	+	+++	+++	+	+	+
Parkinsonian gait	++	+	+	+	+	+	++	+++	−	−	−
Tremor (extrapyramidal)	−	−	−	−	+	−	−	−	−	−	−
Hypokinetic speech	++	+	+++	+	+	−	+++	+++	+	+	−
Response to levodopa	−	−	−	−	++	−	−	++	NA	NA	NA
Ocular dysmetria	+	−	−	−	−	−	−	−	+	−	+
Gait ataxia	+	−	−	++	−	−	−	−	+++	++	+++
Limb ataxia	+	−	+	+	−	−	−	+	+++	++	+++
Ataxic speech	+	−	+	++	−	+	++	−	++	++	+++
Spastic speech	−	++	++	+	−	+	++	−	+	+	+
Extensor plantar signs	−	−	−	−	−	+	+	+	−	+	−
Postural hypotension	++	++	+	−	+	++	+++	++	+++	+	+++
Urinary incontinence	+++	+++	+	−	−	− (Stool +)	++	++	+++	++	++
MRI atrophy						NA	NA				
Cerebral	−	−	−	−	+			−	−	−	−
Basal ganglia	−	−	−	−	+			++	−	−	−
Cerebellar	−	−	−	+	−			+	++	++	+
Brainstem	+	−	+	+	−			+	+++	+++	+

Scales: − = none; + = mild; ++ = moderate; +++ = marked; NA = not available; MRI = magnetic resonance imaging.

sion to MSA, and at post mortem they show neuropathological changes typical of MSA.[1–4]

At present, no method is available to determine whether individual sOPCA patients will progress to develop MSA.[12] The diagnosis of MSA carries a more grave prognosis than sOPCA, hence a means of predicting the evolution of sOPCA into MSA would be useful clinically. In addition, although MSA patients clinically affected by parkinsonism, autonomic dysfunction, and cerebellar manifestations show at post mortem degenerative changes in the brainstem, cerebellum, nigrostriatal connections, and spinal cord, the degree of involvement of these individual structures in patients with principally cerebellar or parkinsonian features is not known.

In a previous investigation, we used (±)-[11C]dihydrotetrabenazine [(±)-[11C]DTBZ], a new ligand for the type 2 vesicular monoamine transporter (VMAT2), with positron emission tomography (PET) to examine the density of striatal monoaminergic presynaptic terminals in MSA compared with sOPCA patients and normal controls.[16] Mean ligand binding was significantly decreased in the striatum of 4 MSA patients and marginally diminished in the striatum of 8 sOPCA patients compared with 9 normal controls. These findings demonstrated severe nigrostriatal pathology in MSA and suggested asymptomatic involvement in the sOPCA group, indicating that some of the

sOPCA patients may later develop parkinsonian features. The MSA group was too small to compare differences in binding in the MSA-P compared with the MSA-C patients studied.

We initiated the current investigation to examine a larger cohort of MSA patients to determine whether striatal monoaminergic presynaptic terminals are differentially affected in MSA-P compared with MSA-C patients and to compare the findings in these two groups with a group of sOPCA patients and a group of similarly aged normal controls. Recently, our radiochemists succeeded in separating the active (+) enantiomer from the inactive (−) enantiomer of DTBZ. In this study we used the (+) enantiomer of [11C]DTBZ, effectively doubling the VMAT2 signal previously obtained with the (±) ligand.[16] A preliminary version of this study has been published in abstract form.[43]

Subjects and Methods

Patient Groups and Normal Subjects

The Institutional Review Board of the University of Michigan approved this investigation. We obtained informed consent from all participants. We studied 7 normal control subjects (all men; age, 57 ± 13 years; mean ± SD), 8 patients with MSA-P (6 men and 2 women; 65 ± 10 years), 8 with MSA-C (3 men and 5 women; 62 ± 5 years), and 6 with sOPCA (2 men and 4 women; 54 ± 18 years) (Table 1). The controls were selected to have age distributions similar

12 MSA-C	13 MSA-C	14 MSA-C	15 MSA-C	16 MSA-C	17 sOPCA	18 sOPCA	19 sOPCA	20 sOPCA	21 sOPCA	22 sOPCA
M	F	F	M	F	F	F	M	M	F	F
63	62	67	67	53	52	39	73	76	28	55
1.5	5	3	2	8	4	3	3	3	17	6
−	+	+	+	−	−	−	−	−	−	−
+	+++	+	+	−	−	−	−	−	−	−
+	+++	+	++	−	−	−	−	−	−	−
−	+++	−	−	+	−	−	−	−	−	−
+	−	+	−	+	+	−	−	−	−	−
NA	+	NA	NA	NA	NA	−	NA	NA	NA	−
+	+	+	+	++	+	+	+	++	++	+++
++	++	+	++	++	+++	++	++	++	++	+++
+	+	−	++	++	+	++	+	+	+++	++
++	++	+	++	+++	++	++	+	−	+	++
−	+	−	−	+	+	NA	−	−	−	++
−	+	−	−	+	−	−	−	+	−	−
+	+	++	+	+++	−	−	−	−	−	−
−	+	+++	+	++	−	−	−	−	−	−
		NA	NA				NA			
−	−			−	−	−		−	−	−
−	−			−	−	−		−	−	−
+	+			++	+++	+++		+++	+++	+
++	+			++	+	+		+	+	++

to those of the patient groups. The controls had no history of neurological disorders and no abnormalities on general physical and neurological examinations. None of the patients or controls were receiving centrally active medications that could influence striatal VMAT2 binding.[16,44]

We classified the patients with MSA according to the presenting feature (parkinsonism or cerebellar ataxia), and in these patients the presenting feature was also the more severe feature (see Table 1). The diagnosis of MSA-P was based on the demonstration of a parkinsonian syndrome in combination with (1) autonomic failure in 5 patients and (2) autonomic failure and cerebellar ataxia in 3. The diagnosis of MSA-C was based on the finding of cerebellar ataxia with (1) autonomic failure in 5 patients, (2) autonomic failure and parkinsonism in 2, and (3) parkinsonism without autonomic failure in 1. The diagnosis of a parkinsonian syndrome required at least two of the following: bradykinesia, rigidity, tremor, or hypokinetic dysarthria, all poorly responsive or unresponsive to levodopa. The diagnosis of cerebellar dysfunction required at least two of the following: gait ataxia, limb ataxia, ataxic dysarthria, or sustained gaze-evoked nystagmus, in the absence of other disorders that could cause progressive ataxia such as sensory loss, medications, toxins, cerebellar neoplasms, paraneoplastic cerebellar degeneration, or multiple sclerosis. The diagnosis of autonomic failure required postural hypotension or urinary incontinence, the latter not caused by either outflow obstruction or abnormal bladder suspension. Although all male patients studied had erectile dysfunction, this was not included in the criteria because of the many possible causes of this symptom. The criteria for postural hypotension were an orthostatic drop of 30 mm or more in systolic blood pressure and 20 mm or more

in diastolic blood pressure with an increase in heart rate of no more than 10 beats per minute[45] in the absence of medication, dehydration, or medical disorders that might cause this finding.[46] We measured blood pressure and pulse with the patient supine and again 2 to 3 minutes after the patient had assumed a standing position.

We studied 6 patients with sOPCA. The diagnosis was based on the finding of a sporadically occurring progressive deterioration of cerebellar function manifested by at least two of the following: gait ataxia, limb ataxia, ataxic dysarthria, or sustained gaze-evoked nystagmus, in the absence of other disorders that could cause progressive ataxia such as sensory loss, medications, toxins, cerebellar neoplasms, paraneoplastic cerebellar degeneration, or multiple sclerosis. To ensure that the disorder was sporadic, we took a full family history, including details concerning the parents, grandparents, aunts, uncles, siblings, and children. The diagnosis of sOPCA was assisted by finding cerebellar atrophy in magnetic resonance (MR) scans in all 6 patients (see Table 1), although this finding was not required as sOPCA can occur without demonstrable atrophy in anatomical imaging studies.[47]

Patient Evaluations

We evaluated all patients with a detailed history, physical examination, neurological examination, laboratory tests to exclude other diseases, and MR imaging to exclude demyelinative disease and structural abnormalities. Speech was evaluated as described previously.[48,49] We obtained a complete blood count, serum profiles of hepatic and renal function, a serological test for syphilis, serum levels of vitamins E, B_{12}, and folic acid, and studies of thyroid function. In patients

with symptoms for fewer than 3 years, we searched for an occult malignancy with breast and pelvic examinations in women, prostate examination in men, acid phosphatase and prostate-specific antigen levels, anti-Purkinje cell antibodies in blood samples, stool guaiac tests for occult blood, and chest x-rays. The neurological examinations were conducted by a neurologist (S.G.) and the speech evaluations by a speech pathologist (K.J.K.), both of whom were blinded to the PET data. Based on the examinations, the parkinsonian and cerebellar features were graded on a scale of $-$ = none, $+$ = mild, $++$ = moderate, and $+++$ = severe (see Table 1). The autonomic features were graded on the same scale based on the history (for urinary incontinence) and the examination (for postural hypotension).

Magnetic Resonance Imaging

Full sets of MR scans were available to review for 17 of the 22 patients studied. The pulse sequence varied among scans, but essentially all included inversion-recovery sequences with T1-weighted, T2-weighted, and proton-density images. One of the authors (L.J.) who was blinded to patient diagnoses reviewed these scans. He evaluated the severity of atrophy in the cerebral cortex, basal ganglia, cerebellum, and brainstem (mesencephalon, pons, and medulla), using perceptual analysis, taking into account patient ages. The degree of atrophy was graded on a scale of $-$ = none, $+$ = mild, $++$ = moderate, and $+++$ = severe (see Table 1). The results of the evaluations of MR scans were not used for quantitative study.

Positron Emission Tomography Studies

The ligand $(+)$-$[^{11}C]$DTBZ binds to the VMAT2 site, and this site is not regulated by the effects of diseases or medications, including levodopa and numerous other pharmaceuticals.[16,44] Hence medications need not be stopped before PET scanning, and in this study none of the medications taken by the patients or normal controls were discontinued. The subjects were placed on a table where they lay quietly through the scanning procedure, eyes open, ears unoccluded, alert but not speaking. We inserted a catheter in a radial artery for blood sampling and placed radioactive fiducial markers on the scalp to register the dynamic sequence of frames, correcting for motion between frames of the study. We imaged the subjects with a CTI PET Systems (Knoxville, TN) ECAT EXACT-47 PET scanner, which has an intrinsic in-plane resolution of 6.0-mm full-width at half-maximum (FWHM) at the center of the field of view and an axial resolution of 5.0-mm FWHM. The reconstructed resolution was approximately 9.0-mm FWHM. Forty-seven planes with a 3.375-mm center-to-center separation were imaged simultaneously. Attenuation correction was calculated by the standard ellipse method.

We prepared $(+)$-$[^{11}C]$DTBZ by ^{11}C-methylation of α-$(+)$-9-O-desmethyl-DTBZ, with a solid phase–supported system allowing purification and isolation of the product $(+)$-$[^{11}C]$DTBZ without high-performance liquid chromatography purification. We administered intravenously 18 ± 1 mCi of $(+)$-$[^{11}C]$DTBZ containing less than 50 μg of mass of DTBZ. Arterial blood samples were collected every 10 seconds for 2 minutes, then at 2.5, 3, 4, 5, 7.5, 10, 15, 20, 30, 45, and 60 minutes after injection. Samples at 1, 2,

and 3 minutes and all subsequent samples were analyzed for radiolabeled metabolites by using C-18 SEP-PAK chromatography as described previously.[44] We recorded a sequence of 15 scans over 60 minutes from the time of injection.

Pharmacokinetic Analysis

We performed pixel-by-pixel fits to a two-compartment tracer kinetic model, using a weighted integration method. The analysis provided parametric images of total tissue distribution volume (DV) of DTBZ relative to authentic DTBZ in arterial plasma[50] and parametric images of ligand transfer from plasma to brain (K_1), which are highly correlated with flow because the single-pass extraction fraction is greater than 50%.

Volumes of Interest Analysis

For measurements from the caudate nucleus and putamen, we created anatomically configured volumes of interest (VOIs) from parametric images of DV, then placed these VOIs on K_1 images. For measurements from the cerebellar hemispheres, we created anatomically configured VOIs from parametric images of K_1, then placed these VOIs on DV images. VOIs were acquired from the two adjacent axial levels that contained the peak parameter values (K_1 or DV) of each of the three structures (caudate nucleus, putamen, and cerebellar hemispheres). VOIs for these three structures were 0.6, 1.1, and 1.7 ml, respectively. As a comparison, with the PET scanner used, the volume of a resolution element is approximately 0.6 ml. For normalization of data obtained from subcortical structures, VOIs were obtained from the cerebral cortex in 8 to 10 adjacent horizontal slices extending from the lowest slice containing the thalamus up to the first slice in which the cortex could be seen along the entire medial surface, which is above the level of the corpus callosum and ventricles. The cerebral cortex contains few specific binding sites, making it an appropriate region for normalization. We subtracted the cortical DV from the total DV of each VOI before normalization [$(DV_{VOI} - DV_{ctx})/DV_{ctx}$] to remove free and nonspecifically bound tracer and thereby obtain a measure of normalized specific binding, commonly termed binding potential (BP). Note that BP is simply DV_{VOI} normalized to cortex minus 1.

Data Analysis

We compared mean BP and K_1 values in the diagnostic groups (MSA-P, MSA-C, sOPCA, and normal controls) by analysis of covariance (ANCOVA), adjusting for age and sex, for each of three anatomical sites (caudate nucleus, putamen, and cerebellar hemispheres). As the groups had similar although not identical mean ages, age adjustment had a small effect in the ANCOVA. Pairwise group comparisons were conducted using t tests, and p values are presented without corrections for multiple comparisons. Within-group variances were treated as constant in BP analyses, but allowed to differ between groups in K_1 analyses to reflect the inequality of variances observed. We normalized to the whole brain cortex, and made no corrections for the effects of tissue atrophy. We examined the relation between symptom intensity of the parkinsonian features and VMAT2 binding in the caudate nucleus and putamen and between symptom inten-

sity of the cerebellar dysfunction and K_1 values in the cerebellum. Regression models were used to obtain correlations for the above relations on all three patient groups. Pearson correlations were computed on age-adjusted values derived from regressions on age from the pooled sample, including normals.

Results

$[^{11}C]$Dihydrotetrabenazine Specific Binding

For the three patient groups compared with the control group, mean BP values adjusted for age and sex were reduced in the caudate nucleus and putamen in the order MSA-P < MSA-C < sOPCA (Table 2). ANCOVA revealed highly significant differences between groups for the caudate nucleus and putamen, but no differences for the cerebellar hemispheres. Comparison of MSA-P patients with controls showed a 59% lower adjusted mean BP in the caudate nucleus ($p < 0.0001$) and a 71% lower adjusted mean in the putamen ($p < 0.0001$). For MSA-C patients compared with controls, corresponding reductions were 43% in the caudate nucleus ($p = 0.002$) and 55% in the putamen ($p < 0.0001$). Adjusted means for MSA-P and MSA-C groups did not differ significantly, but the MSA-P subjects had 27% lower BP in the caudate nucleus and 35% lower BP in the putamen than the MSA-C subjects. For sOPCA patients, adjusted means were intermediate between those of control and MSA groups in both the caudate nucleus and putamen. Adjusted means for both regions were significantly lower for both MSA groups than for the sOPCA group ($p < 0.05$). Comparison of sOPCA patients with controls showed a statistically significant difference in the putamen ($p = 0.04$) but not in the caudate nucleus ($p = 0.25$).

$[^{11}C]D$ ihydrotetrabenazine Transport (K_I)

ANCOVA revealed significant differences between groups for all three regions (see Table 2). Pairwise group comparisons of MSA-P patients with controls showed a nonsignificant 7% decrease of adjusted mean K_1 in the caudate nucleus and a significant ($p = 0.04$) 12% decrease in the putamen. For MSA-C patients, adjusted mean K_1 values in the caudate nucleus and putamen were the lowest of the four groups, and were significantly different from both normal controls and sOPCA patients ($p < 0.05$). Adjusted means of the MSA-C group were decreased 12% in the caudate nucleus and 16% in the putamen compared with controls. Adjusted means of the MSA-C group were decreased 8% in the caudate nucleus and 8% in the putamen compared with the sOPCA group ($p < 0.05$, for each structure). The MSA-P and MSA-C groups did not differ significantly in adjusted means for the caudate nucleus and putamen. For sOPCA patients, adjusted means in the caudate nucleus and putamen were intermediate between those of the controls and MSA patients. Comparison of sOPCA patients with controls revealed no significant change in the caudate nucleus and a 8% reduction in the putamen ($p = 0.03$). In the cerebellar hemispheres, the adjusted mean values for MSA-P patients were the highest of the patient groups and not significantly different from the control group, the values for MSA-C were the lowest, and the sOPCA group fell between the two MSA groups. Pairwise group comparisons with controls showed a 48% decrease in the adjusted mean for MSA-C patients ($p < 0.0001$) and a 22% decrease for sOPCA patients ($p = 0.03$). The adjusted mean for

Table 2. Comparison of PET Measurements of (+) -$[^{11}C]$Dihydrotetrabenazine Binding Potential (BP) and Normalized Ligand Transport (K_I) in Normal Controls and Patients with Multiple System Atrophy Showing Predominantly Parkinsonian Features (MSA-P), Patients with MSA Manifesting Predominantly Cerebellar Features (MSA-C), and Patients with Sporadic Olivopontocerebellar Atrophy (sOPCA)

Measurement	Controls (n = 7)	MSA-P (n = 8)	MSA-C (n = 8)	sOPCA (n = 6)	p^a
BP					
Caudate nucleus	2.68 ± 0.25	1.11 ± 0.20[b]	1.52 ± 0.19[b]	2.28 ± 0.22[c,d]	<0.0001
Putamen	2.92 ± 0.24	0.85 ± 0.19[b]	1.30 ± 0.19[b]	2.22 ± 0.22[b–d]	<0.0001
Cerebellar hemispheres	0.27 ± 0.05	0.26 ± 0.04	0.24 ± 0.04	0.13 ± 0.04	0.125
K_1 (normalized)					
Caudate nucleus	1.16 ± 0.03	1.08 ± 0.07	1.02 ± 0.03[b]	1.11 ± 0.02[d]	0.019
Putamen	1.28 ± 0.03	1.13 ± 0.06[a]	1.08 ± 0.02[b]	1.18 ± 0.03[b,d]	0.0003
Cerebellar hemispheres	1.19 ± 0.04	1.08 ± 0.08	0.62 ± 0.04[b,c]	0.93 ± 0.10[b,d]	<0.0001

Data are mean ± SE values, adjusted for age and sex. Whole brain cortex was used for both calculating BP and normalizing K_1.

[a]From analysis of covariance model F test of equivalent means in the four groups adjusted for age and sex. Weighted least-squares analysis used for all K_1 variables to adjust for unequal variances of the groups.
[b]Statistically significant difference from controls at level 0.05.
[c]Statistically significant difference from MSA-P at level 0.05.
[d]Statistically significant difference from MSA-C at level 0.05.

the cerebellum in MSA-C patients was 43% lower than in MSA-P patients ($p < 0.0001$).

Magnetic Resonance Imaging
The MR scans revealed mild atrophy of the cerebral cortex in 3 of the 8 MSA-P patients, 1 of the 8 MSA-C patients, and none of the sOPCA group (see Table 1). The basal ganglia showed mild to moderate atrophy in 2 MSA-P patients and no atrophy in any of the other patients. In the cerebellum, atrophy was mild to moderate in 4 of the 8 MSA-P patients, mild to moderate in 7 of the 8 MSA-C patients, and mild to severe in all 6 sOPCA patients. In the brainstem, atrophy was mild to severe in 5 of the 8 MSA-P patients, mild to severe in all 8 MSA-C patients, and mild to moderate in 5 of the 6 sOPCA patients.

Relation of Neurological Dysfunction to BP and K_1
The autonomic dysfunction was approximately equal in severity in the MSA-P compared with the MSA-C groups (Table 3). As expected from the clinical classifications of these patients, the parkinsonian signs were much more severe in MSA-P than in MSA-C patients, and the cerebellar dysfunction was much worse in MSA-C than in MSA-P patients (see Table 3). The severity of cerebellar dysfunction was approximately equal in MSA-C compared with sOPCA patients. Analysis of the three patient groups together revealed a strong negative correlation between the intensity of the parkinsonian features and BP in the caudate nucleus ($r = -0.62$, $p = 0.002$) and putamen ($r = -0.61$, $p = 0.002$). For example, a one-unit increase (for example, from $+$ to $++$) in all five components of the scale yielded a 20% decrease in mean caudate nucleus and putamen BP. Analysis of the three patient groups together also revealed a negative correlation between

Table 3. Severity of Autonomic, Cerebellar, and Parkinsonian Features Grouped by Diagnostic Category

Clinical Feature	MSA-P (n = 8)	MSA-C (n = 8)	sOPCA (n = 6)
Autonomic dysfunction	50.0 ± 33.3	58.3 ± 32.1	0.0 ± 0.0
Cerebellar manifestations	15.6 ± 15.1	57.3 ± 18.1	58.3 ± 15.8
Parkinsonian signs	47.5 ± 22.4	25.0 ± 18.8	1.1 ± 2.7

Data are mean ± SD values. Scores for autonomic dysfunction are based on two measures; hence, the maximum score (two measures with three levels of severity) is 6; cerebellar manifestations are based on four measures; hence, the maximum score is 12; scores for parkinsonian signs are based on 5 measures; hence, the maximum score is 15. The scores were transformed to a uniform scale by dividing the individual scores by the maximum value for that score and multiplying by 100.

MSA-P = multiple system atrophy with parkinsonian features; MSA-C = MSA with cerebellar features; sOPCA = sporadic olivopontocerebellar atrophy.

the intensity of the cerebellar dysfunction and cerebellar K_1 ($r = -0.50$, $p = 0.017$). A one-unit change in each of the four clinical measures of cerebellar function resulted in a 16% decrease in K_1.

Discussion
We found significantly decreased specific binding of $(+)$-$[^{11}C]$DTBZ to striatal monoaminergic presynaptic terminals in the caudate nucleus and putamen in MSA-P and MSA-C patients compared with normal controls. Specific binding was not significantly different between the two patient groups, but the MSA-P subjects had substantially lower BPs in the caudate nucleus and putamen than the MSA-C subjects. We also found a strong negative correlation between the intensity of the parkinsonian features and binding in the caudate nucleus and putamen in all patient groups combined. The present study augments our previous investigation of striatal monoaminergic presynaptic terminals utilizing $(\pm)$-$[^{11}C]$DTBZ in MSA and sOPCA.[16] In that study we found in MSA, compared with controls, significantly reduced BP in the caudate nucleus and putamen, but the patient group was insufficient in size to differentiate changes in MSA-P and MSA-C patients.

A recent study[51] using $(+)$-$[^{11}C]$DTBZ to compare striatal monoaminergic presynaptic binding in Parkinson's disease with normal controls revealed declines in BP of 66% (in the anterior putamen) to 74% (in the posterior putamen) in the Parkinson's disease group. These values are close to the declines in BP of 71% in the putamen in MSA-P and 59% in MSA-C patients in the present investigation. The similar level of decline of BP in the putamen in Parkinson's disease and MSA-P patients suggests that the parkinsonian features of both disorders result predominantly from deficiency of the nigrostriatal projections. The lesser decline of BP in the MSA-C than the MSA-P patients may explain the less intense parkinsonian features of the MSA-C patients. In addition, the prominent cerebellar degeneration in MSA-C patients may mitigate to some extent the clinical expression of parkinsonism. The hypotonia typical of cerebellar disease[52] may counteract the rigidity that usually results from loss of nigrostriatal terminals.

In two previous investigations, $[^{18}F]$fluorodopa was used with PET to study striatal monoaminergic presynaptic terminals in MSA. One study included 10 MSA patients, all of whom had a parkinsonian syndrome with autonomic failure, and 7 of the 10 also had cerebellar ataxia.[53] Mean ligand uptake was reduced in the caudate nucleus by 46% and in the putamen by 62% in comparison to the normal controls. In our MSA-P patients, who are clinically similar to the patients in this study, mean ligand binding was reduced in the caudate nucleus by 59% and in the putamen by 71%

in comparison with the normal controls. A second study also contained 10 patients, but all were described as having OPCA with autonomic failure.[15] Mean ligand uptake was reduced by 11% in the caudate nucleus and 29% in the putamen compared with the controls. In our MSA-C patients, who are similar to the patients in this study, mean ligand binding was decreased by 43% in the caudate nucleus and 55% in the putamen. Hence, our study demonstrated greater reductions of binding in striatal monoaminergic presynaptic terminals than either of the previous investigations. This may be because VMAT2 is decreased in direct proportion to loss of nigrostriatal terminals, whereas compensatory up-regulation occurs in some of the processes responsible for [^{18}F]fluorodopa uptake, which include transporter and dopa decarboxylase activity. An alternate explanation is that the disorder in the patients we studied may be more advanced than those in the other studies.

In the sOPCA group compared with the normal controls, specific binding of (+)-[^{11}C]DTBZ was decreased in the caudate nucleus and putamen, and the difference reached statistical significance in the putamen. These findings supplement our previous study, which showed decreased binding in both the caudate nucleus and putamen, but the change was marginally significant in the caudate nucleus and nonsignificant in the putamen.[16] The results of both studies suggest that at least some of the sOPCA patients examined are developing nigrostriatal pathology and in the future may exhibit symptoms of parkinsonism.

In the current study, mean blood-to-brain transport (K_1) of (+)-[^{11}C]DTBZ was significantly decreased in the putamen of all patient groups compared with normal controls, with a slightly greater but nonsignificant decrease in the MSA-C compared with the MSA-P group, and with the least change in the sOPCA group. The finding of significantly decreased K_1 values in the patient groups reflects diminished cerebral blood flow, most likely from decreased metabolic and perfusion demands caused by loss of nigrostriatal terminals. This is in keeping with our earlier finding of decreased striatal glucose metabolism in MSA and sOPCA.[12] K_1 values in the caudate nucleus and putamen were significantly lower in MSA-C than sOPCA patients, reflecting the more severe striatal degeneration in the MSA-C group.

We found significantly decreased K_1 values in the cerebellar hemispheres of MSA-C and sOPCA groups compared with controls. This finding indicates that cerebellar blood flow is decreased in these groups, probably from decreased metabolic and perfusion demands. In the MR scans, cerebellar atrophy varying from mild to severe was found in almost all MSA-C and sOPCA patients, supporting the notion that tissue atrophy is an important factor in the decreased perfusion. In contrast to the other two patient groups, the MSA-P subjects showed no significant difference from controls in cerebellar K_1 values, suggesting that cerebellar blood flow was, at most, minimally decreased. In accord with this, only mild cerebellar dysfunction was found on clinical examination in this group and only 2 patients had MR evidence of cerebellar atrophy, mild in both cases.

In the present study, we defined VOIs by using the peak site of [^{11}C]DTBZ binding in the striatum visualized in the PET images. The excellent signal obtained in these images is partly the result of high levels of binding in the striatum and low levels in the surrounding structures. This method provides a more reliable means of extracting binding information from the structures of interest than anatomical imaging with MR imaging scans. MR imaging might be helpful in delineating the exact boundaries of structures, but would provide no greater accuracy in assessing binding.

Partial volume effects in this study may have influenced the ligand binding data to some extent. As the sizes of the structures, particularly the caudate nucleus, and the VOIs used are moderately small relative to the resolution of the reconstructed scans, partial volume effects would influence data from both patients and control subjects. It is highly unlikely, however, that the binding estimates could reflect only atrophic changes in the striatum of the patient groups for several reasons. First, only 2 patients had noticeable atrophy in the basal ganglia. With minimal atrophy, partial volume effects should be similar in patients and controls. The observed decreases in VMAT2 binding in MSA-P and MSA-C patients were 59% and 43% in the caudate nucleus and 71% and 55% in the putamen, respectively. These decreases far exceed the changes that could be attributed to differences in partial volume effects between patients and controls. Second, the strategy of centering VOIs on the peak binding within the caudate nucleus and putamen reduces partial volume effects. Third, even if slight atrophy in the MSA patients caused greater partial volume effects in patients than controls and this atrophy slightly affected the PET measurements, this would not invalidate the conclusion that DTBZ binding is decreased and nigrostriatal terminals are lost in MSA. If the putamen were atrophied without loss of nigrostriatal terminals, measured binding would be increased. Even in the presence of atrophy, decreased VOI values lead to the conclusion that DTBZ binding to nigrostriatal terminals is decreased in MSA. With respect to the cerebellum, the decreases in K_1 in the MSA-C and sOPCA groups may be influenced by partial volume effects to a greater extent than the basal ganglia. Atrophy results not only from global cerebellar shrinkage, however, but also from shrinkage between folia. MR imaging–based correction for atrophy in the cerebellum may be helpful in correct-

ing for effects from global shrinkage, but would not be helpful for the effects of atrophy between the folia.

Taken together, the data presented suggest fundamental differences between MSA-P and MSA-C groups. Degeneration of striatal monoaminergic presynaptic terminals is greater in MSA-P than MSA-C patients, as indicated by both lower specific binding in the caudate nucleus and putamen and more intense parkinsonian signs. In contrast, degeneration of cerebellar tissue is greater in MSA-C than MSA-P patients, as shown by lower K_1 values, greater atrophy in MR scans, and more severe signs of cerebellar deficiency. These differences may reflect differential severity of degeneration of nigrostriatal and cerebellar systems in these two forms of MSA, with either relative preservation or slower degenerative changes of the cerebellum in MSA-P and the striatum in MSA-P.

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