Putaminal serotonergic innervation

Monitoring dyskinesia risk in Parkinson disease

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ABSTRACT

Objective: To explore serotonergic innervation in the basal ganglia in relation to levodopa-induced dyskinesia in patients with Parkinson disease (PD).

Methods: A total of 30 patients with PD without dementia or depression were divided into 3 matched groups (dyskinetic, nondyskinetic, and drug-naive) for this study. We acquired 2 PET scans and 3T MRI for each patient using [^{11}C]-3-amino-4-(2-dimethylaminomethylphenylsulfanyl)-benzonitrile (^{11}C-DASB) and *N*-(3-[^{18}F]fluoropropyl)-2-carbomethoxy-3-(4-iodophenyl) nortropane (^{18}F-FP-CIT). Then we analyzed binding potentials of the 2 radiotracers at basal ganglia structures and correlations with clinical variables.

Results: We observed no difference in ^{18}F-FP-CIT binding between dyskinetic and nondyskinetic patients, whereas there were differences in ^{11}C-DASB binding for the caudate and putamen. Binding potential ratios (^{11}C-DASB/^{18}F-FP-CIT) at the putamen, which indicate serotoninergic fiber innervation relative to dopaminergic fiber availability, were highest in the dyskinetic group, followed by the nondyskinetic and drug-naive PD groups. ^{11}C-DASB/^{18}F-FP-CIT ratios at the putamen and pallidum correlated positively with Unified Parkinson's Disease Rating Scale (UPDRS) total scores and duration of PD, and pallidal binding ratio also correlated with the UPDRS motor scores. Ratios were not dependent on dopaminergic medication dosages for any of the regions studied.

Conclusions: Relative serotonergic innervation of the putamen and pallidum increased with clinical PD progression and was highest in patients with established dyskinesia. The serotonin/dopamine transporter ratio might be a potential marker of disease progression and an indicator of risk for levodopa-induced dyskinesia in PD. A prospective evaluation is warranted in the future.
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GLOSSARY

^{11}C-DASB = [^{11}C]-3-amino-4-(2-dimethylaminomethylphenylsulfanyl)-benzonitrile; **^{18}F-FP-CIT** = *N*-(3-[^{18}F]fluoropropyl)-2-carbomethoxy-3-(4-iodophenyl) nortropane; **ANCOVA** = analysis of covariance; **BP** = binding potential; **DAT** = dopamine transporter; **HY** = Hoehn & Yahr; **LID** = levodopa-induced dyskinesia; **MMSE** = Mini-Mental State Examination; **PD** = Parkinson disease; **PNaive** = drug-naive PD group; **PTC** = PD control group; **PDSK** = PD dyskinesia group; **SERT** = serotonin transporter; **TDLED** = total daily levodopa-equivalent dose; **UPDRS** = Unified Parkinson's Disease Rating Scale; **VOI** = volume of interest.

Long-term dopamine replacement therapy is essential in the management of Parkinson disease (PD). After initiation of levodopa treatment, most patients experience a "honeymoon" period with well-controlled motor symptoms; however, approximately one-third of these patients develop levodopa-induced dyskinesia (LID) after 5 years of therapy, and the percentage climbs to 60% of patients after 10 years of therapy.[1] Young age at onset, high levodopa dosage, and longer treatment duration are important risk factors for LID.[1,2] More severe disease with increased dopaminergic cell loss also likely plays a role[3,4] in the development of LID, since it usually appears on the more affected side and patients with longer duration of preexisting PD tend to develop LID earlier after initiation of levodopa therapy.[3,5] These epidemiologic features

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indicate potential disease-related factors, especially degree of PD progression that may contribute to LID development.

Serotonergic fiber sprouting occurs in the striatum in animals with high levels of dopaminergic cell loss in animal models of PD.[6] Serotonergic fibers can convert levodopa into dopamine[7]; thus, in advanced-stage PD serotonergic fibers might be a major source of dopamine release in the striatum, replacing the function of dopaminergic fibers. In parkinsonian animal models and autopsied brain of patients with PD, serotonergic fiber density in the putamen is increased in cases of dyskinesia.[8] Investigators observed serotonergic axon terminal sprouting and dopamine release from its terminal in rodent LID models.[8] Agonists of presynaptically located serotonin receptors 1A/1B, which act as negative feedback controllers of dopamine release from serotonergic terminals, attenuate LID in rodent models.[9] The observation of the potential contribution of serotonergic fibers to dyskinesia in PD was also supported by a study conducted in patients with PD and graft-induced dyskinesias who had undergone nigral fetal cell transplantation.[10] Bupropion, a serotonin 1A/1B receptor agonist, markedly reduced this refractory dyskinesia.[10] Taken together, these observations suggest that serotonergic fibers in the putamen release dopamine in a nonphysiologic manner and may contribute to LID in PD; this hypothesis is confirmed by a recent experiment conducted in patients with PD.[11] To date, there is insufficient evidence using in vivo imaging in patients with PD regarding distribution of serotonergic fiber innervation, its correlation with dopaminergic fiber degeneration, and its relation to LID.

METHODS

Study participants. We enrolled consecutive patients who visited our movement disorders clinic during the study period, basing PD diagnosis on the UK PD Brain Bank Society criteria.[12] The drug-naive PD group (PNaive) consisted of patients with early-stage PD who were first diagnosed by movement disorder specialists and had no prior history of antiparkinsonian drug treatment. The PD control group (PTC) consisted of patients who had never experienced dyskinesias despite 5 or more years of levodopa treatment. The PD dyskinesia group (PDSK) consisted of patients on levodopa treatment and with moderate to severe LID consistently noted by investigators at clinic visits. For all 3 groups, patients were excluded if they had cognitive impairment (Mini-Mental State Examination [MMSE] score ≤24), history of depression or other psychiatric illness, excessive daytime sleepiness or other sleep disorders such as restless leg syndrome or obstructive sleep apneas, neurologic diseases other than PD, or other neurosurgical procedures. We also excluded patients taking antipsychotics, antidepressants, or other drugs affecting the dopamine and serotonin systems (except for antiparkinsonian drugs) due to the possibility of alterations in dopamine and serotonin transporter uptake.

Clinical evaluations for each patient included the Unified Parkinson's Disease Rating Scale (UPDRS), the MMSE, the 30-item Geriatric Depression Scale, and Hoehn & Yahr (HY) staging. We also collected clinical data such as age, sex, duration of PD, daily dosages of dopaminergic drugs, and total daily levodopa-equivalent dose (TDLED, calculated as previously described).[13]

PET imaging. Each patient underwent 2 PET scans (Biograph PET/CT scanner, Siemens Biograph 40 Truepoint PET/CT, Knoxville, TN) on different days after 12 hours off medication. After a bolus injection of 10 mCi *N*-(3-[^{18}F]fluoropropyl)-2-carbomethoxy-3-(4-iodophenyl) nortropane (^{18}F-FP-CIT), patients waited for 2 hours and then underwent 10-minute scans following a previously reported protocol.[14] For the second scan, patients waited for 1 hour after bolus injection of 10 mCi [^{11}C]-3-amino-4-(2-dimethylaminomethylphenylsulfanyl)-benzonitrile (^{11}C-DASB) and then underwent a 30-minute scan. We chose this protocol of ^{11}C-DASB scan[15] because of a report of good agreement between the ratio analysis method using a single static image and the conventional compartmental modeling using dynamic PET data. For each scan, we collected emission data in 3-dimensional mode. After routine corrections for physical effects, individual static images were reconstructed onto a 256 × 256 × 74 matrix with a voxel size of 1.34 × 1.34 × 3 mm^3 using a 3D OSEM PSF reconstruction algorithm (6 iterations and 21 subsets).

MRI acquisition and processing. We acquired high-resolution brain MRI scans (Philips Achieva 3.0T, Best, the Netherlands) for each patient (T1-weighted images with repetition time/echo time = 9.9/4.6, flip angle of 8.0°, 1 mm slice thickness, an image matrix of 224 × 224 × 180, and voxel size of 0.98 × 0.98 × 1 mm^3). We determined the anatomical boundaries of the putamen, caudate nucleus, globus pallidus, thalamus, and cerebellum (used as a reference region) on the T1-weighted MRI of each patient using the FMRIB Integrated Registration and Segmentation Tool (FIRST, FSL v4.0, Oxford University, UK, http://www.fmrib.ox.ac.uk/fsl). Then we coregistered individual PETs and MRIs and conducted spatial normalization of the MRI to the MNI152 template using the SPM8 package (Statistical Parametric Mapping, University College of London, UK).

Ratio analysis. Target-to-reference ratio of uptake after equilibrium provides a measure of binding potential (BP). We used the ratio analysis method to estimate the binding potential (BP_{ratio}) because of reports of good agreement between this method and conventional compartmental modeling using dynamic PET data.[15] For each static image, we divided by the uptake value in the cerebellum that was extracted by applying the corresponding volume of interest (VOI) to the static image and then subtracted by 1, finally yielding the parametric image of

$$BP_{ratio} = \left(\frac{voxel}{cerebellum} - 1\right).$$

We calculated regional BP_{ratio} values in both hemispheres by applying each MRI-based VOI to the parametric images and additionally subtracted regional BP_{ratio} values for anterior and posterior putamen and raphe nucleus regions using the statistical probabilistic anatomy map method.[16,17]

Statistical analysis. For comparisons among the 3 groups in terms of clinical variables and BPs at VOIs, we used the analysis of variance test for continuous variables with post hoc Bonferroni test for subgroup comparisons and the χ^2 test for categorical variables. We analyzed correlations between the BP ratios of DASB/FP-CIT and clinical variables using Pearson correlation analysis and examined the effects of clinical variables on BP ratios using the analysis of covariance (ANCOVA). Significance was set at 0.05 (2-tailed), and we used SPSS software version 19.0 (SPSS Inc., Chicago, IL) for statistical analyses.

Standard protocol approvals, registrations, and patient consents. The Institutional Review Board of Seoul National University Hospital approved this study. All study patients gave informed consent prior to participation in the study, according to the Declaration of Helsinki.

RESULTS

Clinical and demographic characteristics. Thirty patients (10 for each group) enrolled in this study, and there were no dropouts and no adverse events. We did not observe differences in age, sex, and PD onset age among the 3 groups (table 1). Similarly, for the PTC and PDSK groups, there were no differences in PD duration, onset age, and UPDRS part I, II and III scores; however, UPDRS part IV scores and TDLED were higher in the PDSK group.

Dopaminergic fibers in the basal ganglia: ^{18}F-FP-CIT BP analysis. At the caudate, putamen, and globus pallidus, the BP decrease was less severe in the PNaive group than in the PTC and PDSK groups ($p < 0.001$); however, we observed no intergroup difference between the PTC and PDSK groups in all 3 regions (table 2) and no difference in the thalamic BPs among the 3 groups. The estimated BPs in the caudate and putamen correlated negatively with PD duration, HY stage, and total UPDRS score ($p < 0.05$ for all comparisons). There was no correlation between ^{18}F-FP-CIT binding and TDLED in all regions in our patients.

Serotonergic fiber densities in the basal ganglia: ^{11}C-DASB BP analysis. The ^{11}C-DASB BP comparisons are shown in table 3. There was a difference at the caudate and putamen ($p < 0.001$). A reduction was found in the PDSK group compared with the other 2 groups, whereas there was no difference between the PNaive and PTC groups (table 3). At the pallidum, raphe nucleus, and thalamus, there were no group differences in the ^{11}C-DASB BPs. ^{11}C-DASB binding decrease in the caudate, but not in the putamen and pallidum, correlated with PD duration and total UPDRS score ($p < 0.05$ for both). There was no correlation between ^{11}C-DASB binding and TDLED at all regions.

Serotonin to dopaminergic fiber availability estimated by BP ratios. We assessed serotonin to dopaminergic fiber availability by measuring BP ratios of ^{11}C-DASB/^{18}F-FP-CIT. We observed the highest ^{11}C-DASB/^{18}F-FP-CIT ratios in the putamen in the PDSK group, followed by the PTC and PNaive groups (figure 1), and a more obvious linear increase of ^{11}C-DASB/^{18}F-FP-CIT binding ratio at the

Table 1 Clinical characteristics of the patients with Parkinson disease enrolled in this study

Characteristics	Drug-naive	No dyskinesia	Dyskinesia	p Value[a]	p Value[b]	p Value[c]
Age, y	61.3 ± 10.7	61.5 ± 9.2	63.9 ± 7.8	0.787	1.000	1.000
Sex, F/M	4/6	3/7	4/6	0.866[d]		
Age at onset, y	60.3 ± 10.6	54.5 ± 11.4	55.7 ± 9.5	0.441	0.687	1.000
Duration of PD, y	1.1 ± 0.5	7.0 ± 4.9	8.2 ± 2.6	<0.001	0.001	1.000
Hoehn & Yahr stage, 1–5	1.6 ± 0.5	1.8 ± 0.5	2.1 ± 0.4	0.042	0.706	0.470
MMSE score, 0–30	27.1 ± 2.0	27.7 ± 1.4	27.5 ± 2.2	0.774	1.000	1.000
GDS score, 0–30	13.4 ± 6.7	11.4 ± 6.0	13.2 ± 5.5	0.721	1.000	1.000
Total UPDRS score	34.2 ± 12.9	33.4 ± 12.0	51.4 ± 14.1	0.007	1.000	0.014
Part I	7.3 ± 5.1	5.6 ± 3.9	8.2 ± 3.9	0.408	0.969	0.574
Part II	5.7 ± 4.3	7.1 ± 4.0	11.3 ± 5.9	0.040	1.000	0.187
Part III	21.2 ± 6.4	20.4 ± 9.8	23.0 ± 5.9	0.736	1.000	1.000
Part IV	0 ± 0	0.3 ± 0.9	8.8 ± 3.2	<0.001	1.000	<0.001
LED, mg/d	0 ± 0	713.5 ± 158.7	943.7 ± 213.5	<0.001	<0.001	0.007

Abbreviations: GDS = Geriatric Depression Scale; LED = levodopa-equivalent dose; MMSE = Mini-Mental State Examination; PD = Parkinson disease; UPDRS = Unified Parkinson's Disease Rating Scale.
Data are shown as mean ± SD, unless otherwise indicated.
[a] Comparison of 3 groups by the analysis of variance.
[b] Comparison of drug-naive PD and nondyskinesia groups using post hoc Bonferroni analysis, $p < 0.05$.
[c] Comparison of nondyskinesia and dyskinesia groups using post hoc Bonferroni analysis, $p < 0.05$.
[d] Comparison of 3 groups by the χ^2 test.

Table 2 Estimated ^{18}F-FP-CIT binding potentials at volumes of interest

	Drug-naive	No dyskinesia	Dyskinesia	*p* Value[a]	*p* Value[b]	*p* Value[c]	*p* Value[d]
Caudate nucleus	4.79 ± 1.48	3.37 ± 0.89	2.70 ± 1.10	<0.001	0.001	0.243	<0.001
Putamen	3.95 ± 1.61	2.30 ± 0.57	1.63 ± 0.52	<0.001	<0.001	0.139	<0.001
Anterior	4.15 ± 1.33	2.42 ± 0.69	1.84 ± 0.67	<0.001	<0.001	0.169	<0.001
Posterior	3.05 ± 1.78	1.66 ± 0.51	1.10 ± 0.33	<0.001	<0.001	0.320	<0.001
Globus pallidus	1.88 ± 0.39	1.40 ± 0.20	1.32 ± 0.26	<0.001	<0.001	1.000	<0.001
Thalamus	0.59 ± 0.11	0.58 ± 0.09	0.57 ± 0.09	0.723	1.000	1.000	1.000

Abbreviations: ^{18}F-FP-CIT = *N*-(3-[^{18}F]fluoropropyl)-2-carbomethoxy-3-(4-iodophenyl) nortropane; ANOVA = analysis of variance; PD = Parkinson disease. Data are shown as mean ± SD.
[a] Comparison of the 3 groups using ANOVA.
[b] Comparison of drug-naive PD and nondyskinesia groups using post hoc Bonferroni analysis, $p < 0.05$.
[c] Comparison of nondyskinesia and dyskinesia groups using post hoc Bonferroni analysis, $p < 0.05$.
[d] Comparison of drug-naive PD and dyskinesia groups using post hoc Bonferroni analysis, $p < 0.05$.

posterior putamen compared with the anterior putamen (figure e-1 on the *Neurology*® Web site at Neurology.org). For the pallidum, we observed group differences ($p < 0.001$), but not between the PTC and PDSK groups. There were no differences between groups in binding ratios (figure 1) for the caudate and thalamus.

Correlations of ^{11}C-DASB/^{18}F-FP-CIT binding ratios with PD severity and dopaminergic medication dosages. Because we observed overlaps and linear decreases of both tracer bindings in the PTC and PDSK groups, we wished to clarify whether increasing serotonin to dopaminergic fiber availability was secondary to PD progression or if it was specifically related to the dyskinesia.

We observed a linear correlation for ^{11}C-DASB/^{18}F-FP-CIT with total UPDRS scores (figure 2A) at the putamen and pallidum, but for motor UPDRS scores we only observed a correlation with pallidal ^{11}C-DASB/^{18}F-FP-CIT binding ratios (figure 2B). ^{11}C-DASB/^{18}F-FP-CIT binding ratios at the putamen (figure e-2) and pallidum also correlated with the duration of PD ($r = 0.4560$, $p < 0.001$; $r = 0.5171$, $p < 0.001$; and $r = 0.2879$, $p = 0.026$ for anterior putamen, posterior putamen, and pallidum, respectively). Unlike PD duration and severity, we did not see a correlation of TDLED with increased ^{11}C-DASB/^{18}F-FP-CIT ratios in the putamen (figure e-3) or pallidum ($r = 0.1648$, $p = 0.310$ and $r = 0.0874$, $p < 0.592$ for anterior and posterior putamen, respectively, and $r = 0.1801$, $p = 0.266$ for pallidum). We also observed the 3-group difference in putaminal ^{11}C-DASB/^{18}F-FP-CIT ratios by ANCOVA analysis with total and motor UPDRS scores and PD duration as covariates ($p = 0.012$).

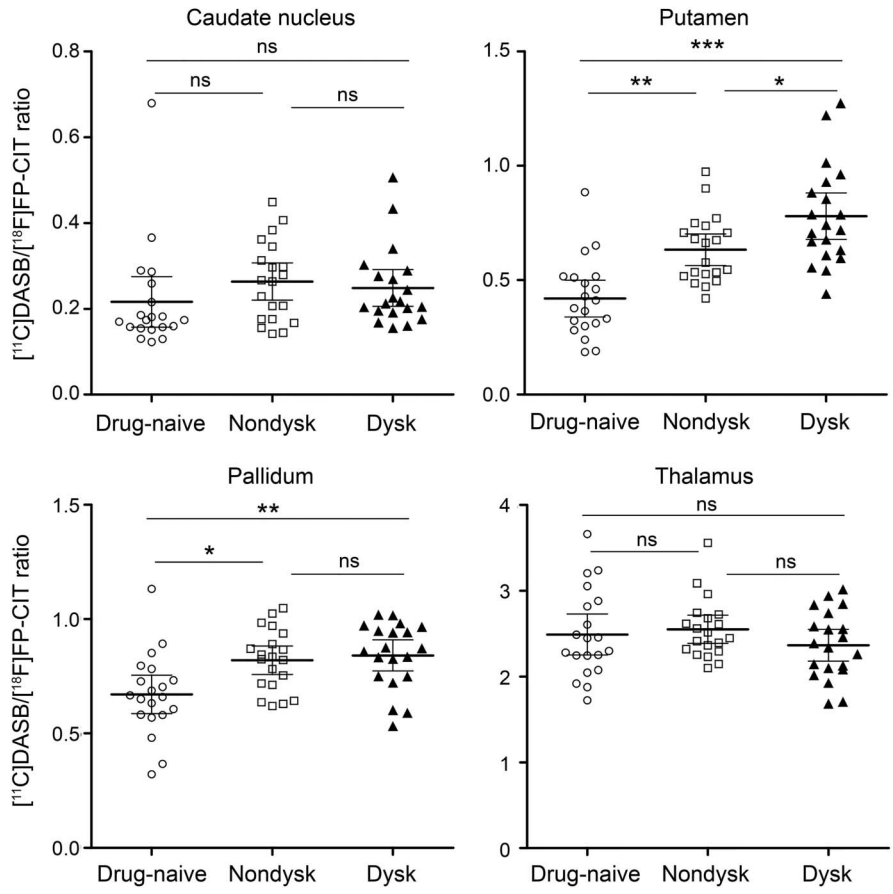
DISCUSSION In this study, we conducted in vivo investigation of serotonin to dopaminergic fiber availabilities in basal ganglia structures correlated with LID in PD. Results of scans with the 2 tracers, ^{11}C-DASB and ^{18}F-FP-CIT, show functional loss of nerve terminals expressing serotonin transporters (SERT)

Table 3 Estimated ^{11}C-DASB binding potentials at volumes of interest

	Drug-naive	No dyskinesia	Dyskinesia	*p* Value[a]	*p* Value[b]	*p* Value[c]	*p* Value[d]
Caudate nucleus	0.92 ± 0.22	0.83 ± 0.20	0.61 ± 0.15	<0.001	0.508	0.002	<0.001
Putamen	1.46 ± 0.27	1.39 ± 0.18	1.20 ± 0.24	0.003	0.975	0.043	0.003
Anterior	1.58 ± 0.27	1.40 ± 0.21	1.20 ± 0.23	<0.001	0.063	0.028	<0.001
Posterior	1.43 ± 0.26	1.34 ± 0.21	1.13 ± 0.23	<0.001	0.600	0.019	<0.001
Globus pallidus	1.21 ± 0.25	1.14 ± 0.21	1.09 ± 0.22	0.232	0.979	1.000	0.270
Thalamus	1.45 ± 0.32	1.46 ± 0.19	1.32 ± 0.24	0.172	1.000	0.282	0.349
Raphe nucleus	3.09 ± 0.62	3.34 ± 0.64	3.24 ± 0.90	0.740	1.000	1.000	

Abbreviations: ^{11}C-DASB = [^{11}C]-3-amino-4-(2-dimethylaminomethylphenylsulfanyl)-benzonitril; ANOVA = analysis of variance; PD = Parkinson disease. Data are shown as mean ± SD.
[a] Comparison of 3 groups using ANOVA.
[b] Comparison of drug-naive PD and nondyskinesia groups using post hoc Bonferroni analysis, $p < 0.05$.
[c] Comparison of nondyskinesia and dyskinesia groups using post hoc Bonferroni analysis, $p < 0.05$.
[d] Comparison of drug-naive PD and dyskinesia groups using post hoc Bonferroni analysis, $p < 0.05$.

Figure 1 ^{11}C-DASB/^{18}F-FP-CIT binding potential ratios in the caudate nucleus, putamen, globus pallidus, and thalamus



Comparisons of [^{11}C]-3-amino-4-(2-dimethylaminomethylphenylsulfanyl)-benzonitrile (^{11}C-DASB)/*N*-(3-[^{18}F]fluoropropyl)-2-carbomethoxy-3-(4-iodophenyl) nortropane (^{18}F-FP-CIT) binding potential ratios among patients with drug-naive, nondyskinetic (Nondysk), and dyskinetic (Dysk) Parkinson disease. ns = no difference; *$p < 0.05$; **$p < 0.01$; ***$p < 0.001$.

and dopamine transporters (DAT)in the corresponding regions. Serotoninergic fibers have the potential to convert L-dopa into dopamine; thus, in advanced PD where substantial dopaminergic fiber degeneration occurs, these fibers may be a major source of dopamine release in the basal ganglia.[8]

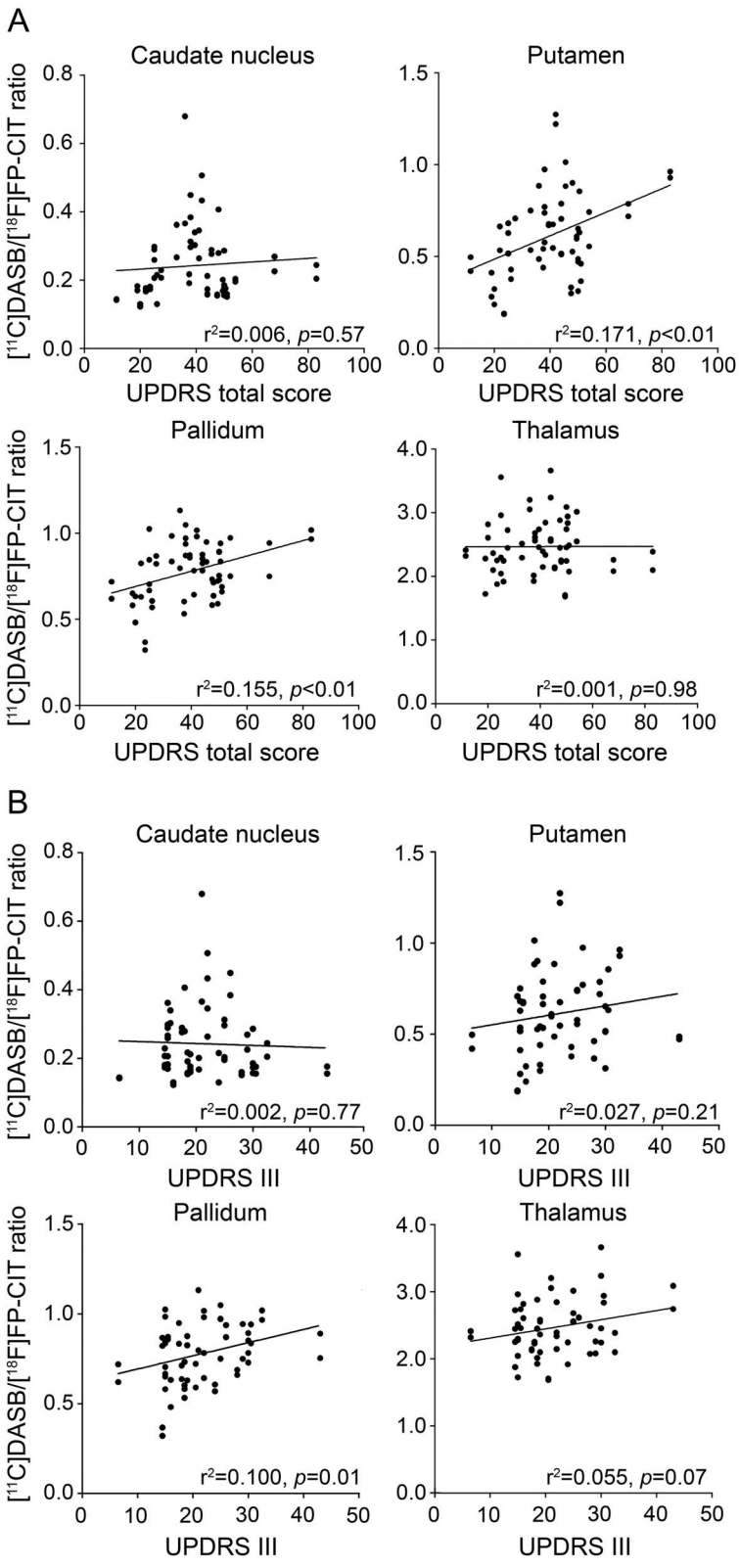
In our patients, we observed decreases of both tracer uptakes with PD disease progression, but each had a distinct topographical pattern. At the caudate nucleus, ^{11}C-DASB binding loss correlated with severity and duration of PD. However, at the putamen, we observed decreased ^{11}C-DASB binding but without a correlation with disease progression. There was no decrease in the thalamus and raphe nucleus despite advanced PD in our patients.

In the raphe nucleus, SERT are expressed mainly on the cell bodies[18]; thus, serotonergic degeneration in PD occurs at the axonal fibers not at the cell bodies, especially for caudate innervation. In previous human PD studies, investigators reported that raphe SERT binding decreased at very advanced PD stages and rostral to caudal degeneration in the serotonergic system in PD.[19,20] Results of in vivo imaging studies, including ours, are consistent with human autopsied brain data showing profound loss in serotonergic markers in the caudate compared with the putamen in PD.[21] This serotonergic loss does not seem to occur in the very early stage of PD.[22]

Unlike SERT binding, DAT binding loss correlates with PD duration and UPDRS scores for both caudate and putamen. Our 2 treated PD groups were matched for disease duration, HY stage, and motor UPDRS score, and we did not observe differences in decreased DAT binding in the striatal region in the PDSK group compared with the PTC group.

To evaluate the contribution of serotonergic fibers to dopaminergic fiber degeneration, we estimated relative SERT availability as expressed in ^{11}C-DASB to ^{18}F-FP-CIT BP ratios. For the putamen, we observed high BP ratios in the PDSK group compared with the PTC group (mean value of ^{11}C-DASB/^{18}F-FP-CIT was 0.42 in the PNaive group, similar to the previously reported value under normal conditions).[10] The mean value of ^{11}C-DASB/^{18}F-FP-CIT in the PDSK

Figure 2 Correlations between ^{11}C-DASB/^{18}F-FP-CIT binding potential ratios and UPDRS scores



Scattergrams of [^{11}C]-3-amino-4-(2-dimethylaminomethylphenylsulfanyl)-benzonitrile (^{11}C-DASB)/*N*-(3-[^{18}F]fluoropropyl)-2-carbomethoxy-3-(4-iodophenyl) nortropane (^{18}F-FP-CIT) binding potential ratios according to Unified Parkinson's Disease Rating Scale (UPDRS) total scores (A) and UPDRS motor scores (B). Correlation coefficients and corresponding *p* values for correlation analysis of the binding potential ratios and UPDRS scores are shown in each scattergram.

group was as high as 0.78, which is similar to that reported in a case with graft-induced dyskinesia.[10] In the PTC group, the value was intermediate. The high ^{11}C-DASB/^{18}F-FP-CIT ratio in the putamen in patients with advanced PD was possibly due to disproportionately severe degeneration in the nigrostriatal dopaminergic fibers despite serotonergic degeneration. On the contrary, the high ratio for the putamen might be induced by relative sparing or maladaptive sprouting of serotonergic fibers in the putamen as compensation for dopaminergic fiber loss.[8] In our study, the pallidal ratio was also high in the 2 treated groups compared with the drug-naive group. The pallidal ^{11}C-DASB/^{18}F-FP-CIT ratio correlated with motor UPDRS scores and PD duration, although we observed no difference in the pallidal ratio between the dyskinetic and nondyskinetic patients. Thus, pallidal serotonergic fibers do not seem to be specifically responsible for LID; however, they are likely to be more strongly correlated with motor severity of PD.

Analysis of BP ratios throughout the various stages and disease durations in our patients showed that increasing serotonergic availability in the putamen seems to be an intrinsic condition occurring in PD brain with disease progression. Serotonergic to dopaminergic fiber availability in the putamen increased as the disability and disease duration increased. Thus, blockade of serotonergic fibers to attenuate LID might not always be helpful with respect to parkinsonian motor symptoms in patients with advanced-stage PD because of the possibility of reducing the antiparkinsonian benefit of levodopa. Results of clinical trials and other studies showed that 5-HT$_{1A}$ agonists impaired levodopa antiparkinsonian action even though they had antidyskinetic effects.[23–28] Two phase III trials of sarizotan failed to show antidyskinetic effects using a low dose to avoid loss of the antiparkinsonian benefit of levodopa.[29,30]

The presynaptic component is important in LID. Animal experiments provided convincing evidence that attenuation of serotonergic fibers or blockade of dopamine release from serotonergic fibers can reduce LID by restoring nonphysiologic dopamine release into the synaptic space in patients with LID.[31–35] In a human study, enhanced synaptic dopamine release as well as LID were partially attenuated following administration of buspirone, an agonist of 5-HT$_{1A}$, in patients with LID. In this study, ^{11}C-DASB PET imaging confirmed increased serotonergic fiber binding in patients with LID.[11] From the present study's results, we suggest that serotonergic fiber innervation might be a marker of disease progression and that abnormal increases in serotonin to dopaminergic availability in the putamen might be a sign that patients with PD are in advanced stages and prone to development of LID. Note that there was

high variability in the ^{11}C-DASB/^{18}F-FP-CIT ratios in our patients; thus, subsequent plastic changes following or accompanying serotonergic innervation would be needed in this process at presynaptic and/or postsynaptic levels.[36,37] There is supporting evidence that serotonergic fibers play key roles in initiating cascades of synaptic plasticity in PD animal models with LID.[38] Currently, the precise mechanism of serotonergic fiber innervation and interactions with nondopaminergic modulators as well as dopamine remains largely unknown. We also need to elucidate whether nondopaminergic fibers also contribute to the serotonergic fiber innervation and plastic changes in signal transduction. Furthermore, it is unknown whether the serotonergic fiber innervation occurs in parkinsonian disorders other than PD in which dopaminergic degeneration occurs. After resolving these issues, investigation will be needed to determine whether early modulation of serotonergic fibers in patients with PD can prevent development of LID.

Although we systematically analyzed basal ganglia serotoninergic fiber changes in dyskinetic vs nondyskinetic patients with PD, these results should be interpreted with caution. The sample size was rather small; thus, we enrolled patients matched for disease onset, level of motor disability, and duration of PD. Because this was not longitudinal data, we cannot make conclusions about causal relationships. The ^{18}F-FP-CIT ligand used in this study has cross-affinity to SERT, although this is quite weak compared with the β-CIT.[39] Thus, we may have underestimated DAT decreases from ^{18}F-FP-CIT binding, especially in serotonin-rich areas, and the actual ^{11}C-DASB to ^{18}F-FP-CIT ratio in the caudate nucleus compared with the putamen may be higher than shown in our results.

Despite these limitations, using in vivo imaging, we observed serotoninergic fiber changes in the basal ganglia in patients with PD. Relative increases in the availability of serotonergic fibers compared with dopaminergic fibers might be a predictor of future risk of dyskinesia and might be a more sensitive marker of disease severity than clinical variables such as disease duration or UPDRS scores. To confirm this finding, further longitudinal studies with prospective evaluation of serotonergic fiber changes in the basal ganglia are warranted.

AUTHOR CONTRIBUTIONS

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DISCLOSURE

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