Frequency of Cholinergic and Caudate Nucleus Dopaminergic Deficits Across the Predemented Cognitive Spectrum of Parkinson Disease and Evidence of Interaction Effects

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**IMPORTANCE** Little is known about the relative contributions of multisystem degenerative processes across the spectrum of predemented cognitive decline in Parkinson disease (PD).

**OBJECTIVE** To investigate the relative frequency of caudate nucleus dopaminergic and forebrain cholinergic deficits across a spectrum of cognitively impaired patients with PD to explore their relative, individual, and combined contributions to cognitive impairment in PD.

**DESIGN, SETTING, AND PARTICIPANTS** A cross-sectional study at an academic movement disorders clinic that included a predominantly nondemented cohort of 143 patients with PD. The mean (SD) age of patients was 65.5 (7.4) years and the mean (SD) Hoehn and Yahr stage was 2.4 (0.6).

**MAIN OUTCOMES AND MEASURES** Binary classification of carbon 11–labeled \(^{[11C]}\)PMP acetylcholinesterase and caudate nucleus \(^{[11C]}\)DTBZ monoaminergic positron-emission tomography imaging based on normative data. The frequency of significant degenerative processes based on normative values was determined for consecutive intervals of cognitive impairment, ranging from no or minimal \((z > −0.5)\) to more severe \((z ≤ −2)\) cognitive impairment.

**RESULTS** Across the spectrum from minimal \((z > −0.5)\) to more severe \((z ≤ −2)\) global cognitive impairment scores, caudate nucleus dopaminergic denervation was relatively frequent in individuals with minimal or no cognitive changes (51.1%) and increased in patients with more severe cognitive impairments \((χ^2 = 12.8; P = .01)\). Cortical cholinergic denervation frequency increased monotonically with increasing cognitive impairment from 24.7% \((z > −0.5)\) to 85.7% \((z ≤ −2)\); \(χ^2 = 23.2; P = .001)\). Eighty-seven percent of patients with neocortical cholinergic deficits had caudate nucleus dopaminergic deficits. Multiple regression analysis \((F = 7.51; P < .001)\) showed both independent cognitive predictions for caudate nucleus dopaminergic \((F = 7.25; P = .008)\) and cortical cholinergic \((F = 7.50; P = .007)\) degenerations as well as interaction effects \((F = 5.40; P = .02)\).

**CONCLUSIONS AND RELEVANCE** Cortical cholinergic denervation is a major neurodegeneration associated with progressive declines across the spectrum of cognitive impairment in PD and typically occurs in the context of significant caudate nucleus dopaminergic denervation. Our findings imply that dopaminergic and cholinergic degenerations exhibit both independent and interactive contributions to cognitive impairment in PD.
Clinical studies of the heterogeneous nature of cognitive impairments in Parkinson disease (PD) have focused on disentangling and characterizing individual variation in deficits, differences in treatment responses, and risk estimations for conversion to PD dementia.1-2 An intriguing dual syndrome hypothesis was proposed to explain the heterogeneity of cognitive changes in PD.2 This model posits that the high frequency of frontostriatal executive dysfunction relates to common dopaminergic deficits and that the development of dementia is associated with more widespread cortical changes secondary to additional pathologies, including cholinergic deficits.2,3 In vivo neuroimaging studies provide some support for this hypothesis. Although dopaminergic denervation affects specific cognitive functions in PD, striatal and limbicfrontal dopaminergic changes are also present in nondemented patients with PD and their presence is not sufficient to explain the development of dementia in PD.3 In contrast, greater cholinergic denervation is shown consistently in PD dementia compared with PD.3,4

In addition to our study, other studies have previously reported on dopaminergic and cholinergic associations with cognitive impairment in PD.3,5 The purpose of the current study was to directly compare the relative frequency and potential for interactions between these 2 neurodegenerations across the mostly nondemented cognitive spectrum in PD. To determine the frequency of each of the different pathologies, we used normative data to dichotomize significant deficits. Although profound putaminal nigrostriatal denervation is a defining feature of patients with PD with essentially no overlap with putaminal dopaminergic denervation in normal control participants, denervation of the caudate nucleus, which has prominent nonmotor cognitive connections, is less severe and occurs in a range with partial overlap between innervation levels seen in PD and those of normal control participants.6,7 We also investigated possible interactive cognitive effects between these 2 system degenerations in PD. We hypothesized that unlike more common dopaminergic deficits, frequency of cholinergic denervation is low in patients with PD with minimal cognitive changes but very prevalent in individuals with more severe cognitive impairment. Furthermore, we hypothesized that combined dopaminergic and cholinergic changes are not merely additive but also have interactive effects on cognitive performance in PD.

Each patient underwent a detailed cognitive examination following an approach previously reported to characterize cognitive impairment in PD.11 These tests included the following measures: verbal memory on the California Verbal Learning Test;2 attention/psychomotor speed as absolute time on the Stroop 1 Test;3 executive/reasoning functions on the Wechsler Adult Intelligence Scale III Picture Arrangement Test;3 and the Delis-Kaplan Executive Function System Sorting Test;4 visuospatial function in the Benton Judgment of Line Orientation Test.6 Composite z scores were calculated for these different cognitive domains based on normative data. A global composite cognitive z score was calculated as the mean of the domain z scores.

To study the relative contributions of different degenerative processes, we stratified our cohort by severity of cognitive impairment. The magnitude of cognitive impairment was fractionated using categories of consecutive 0.5 z score ranges of global composite z scores. The study groups ranged from no or minimal global cognitive impairments (z > −0.5), mild to moderate cognitive impairments (−0.5 to −1.0, −1.0 to −1.5, and −1.5 to −2), and more severe cognitive impairment (z ≤ −2).

The study was approved by and study procedures were followed in accordance with the ethical standards of the institutional review board of the University of Michigan. Written informed consent was obtained from all participants.

Imaging Techniques

All participants underwent brain magnetic resonance imaging and carbon 11-labeled (11C) methyl-4-piperidinyl propionate (PMP) acetylcholinesterase (AChE) and [11C]dihydrotetrabenzine (DTBZ) vesicular monoamine transporter type 2 positron emission tomography (PET) except for 1 patient where the DTBZ PET scan failed for technical reasons. All DTBZ PET scans showed evidence of nigrostriatal degeneration. Magnetic resonance imaging was performed on a 3-Tesla Achieva system (Philips) and PET imaging was performed in 3-dimensional imaging mode with an ECAT Exact HR+ tomograph (Siemens Molecular Imaging Inc), as previously reported.5

The [11C]DTBZ and [11C]PMP were prepared as previously described.17 Dynamic PET scanning performed for 70 minutes using a bolus dose of 15 mCi was used for [11C]PMP. A 60-minute bolus/infusion protocol was used for [11C]DTBZ (15 mCi).18

Image Analysis

Imaging registration was performed as previously reported.5 Interactive Data Language image analysis software (Research Systems Inc) was used to manually trace volumes of interest on magnetic resonance images to include the thalamus and neostriatum (caudate nucleus and putamen) of each hemisphere. Total neocortical volume of interest was defined using semiautomated threshold delineation of the cortical gray matter signal on the magnetic resonance imaging scan. The AChE [11C]PMP hydrolysis rates (k3) were estimated using the striatal volume as the tissue reference.19 The AChE PET imaging as-

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sessed the 2 major brain cholinergic projection systems with cortical uptake reflecting largely basal forebrain and thalamic uptake and principally reflecting pedunculopontine nucleus/complex integrity.

The [11C]DTBZ caudate nucleus distribution of volume ratios were estimated using the Logan plot graphical analysis method using cortical reference tissue. The mean (SD) caudate nucleus vesicular monoamine transporter type 2 distribution volume ratio was 1.89 (0.37) (range, 1.23–3.49). The mean (SD) forebrain neocortical and pedunculopontine nucleus/complex-thalamic AChE hydrolysis rates were 0.0237 (0.0028) (range, 0.0167–0.0333 minute⁻¹) and 0.0546 (0.0055) (range, 0.0377–0.0681 minute⁻¹), respectively.

### Data Analysis

The primary purpose of this study was to directly extend our previous work and investigate the relative frequency of binary defined (present or absent) substantial caudate nucleus dopaminergic and cholinergic degenerative processes at different stages of cognitive impairment in PD in a more expanded patient population. Specifically, we estimated the frequency of each degenerative process in each global cognitive subgroup by classifying each participant as normal or abnormal for cholinergic forebrain or caudate dopaminergic nerve terminals, as measured by the 2 different PET probes. Because of the significant overlap of cholinergic innervation seen between patients with PD and healthy control participants, we classified AChE PET scans as either below or within normal range. Abnormal neocortical and thalamic cholinergic innervation was based on a 5th percentile cut off from non-PD elderly normative data (n = 29; mean [SD] age, 66.8 [10.9] years) comparable with the current patient cohort.

### Results

Relative Frequency of Dopaminergic and Cholinergic Deficits

Table 1 provides a summary of the relative frequencies of significant caudate nucleus dopaminergic denervation and significant neocortical and thalamic AChE deficits status in each cognitive severity subgroup for the total patient cohort (n = 143). Significant caudate nucleus dopaminergic denervation was present in about two-thirds of all patients while cortical cholinergic denervation was present in only about one-third of all patients in this predominantly nondemented cohort. Thalamic cholinergic denervation was present in about one-sixth of patients (Table 1).

Combined Dopaminergic and Cholinergic Deficits

Table 2 shows the relative frequencies of combinations of significant caudate nucleus dopaminergic and neocortical cholinergic deficits in the different cognitive severity subgroups.

Abbreviations: AChE, acetylcholinesterase; PPN, pedunculopontine nucleus/complex; VMAT2, vesicular monoamine transporter type 2.

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### Table 1. Frequency Listing of Significant Caudate Nucleus Dopaminergic, Neocortical, and Pedunculopontine Nucleus/Complex-Thalamic Cholinergic Deficits in the Total Cohort of Patients and Cognitive Severity Subgroups Based on Global Cognitive z Scores

<table>
<thead>
<tr>
<th>Neurochemical Deficit</th>
<th>z Score Ranges, No. (%)</th>
<th>Statistical Significance, χ²</th>
<th>P Value</th>
</tr>
</thead>
<tbody>
<tr>
<td>Caudate nucleus VMAT2</td>
<td>z ≤ −2 Cognitive Subgroup (n = 7)</td>
<td>5 (71.4)</td>
<td>11 (100)</td>
</tr>
<tr>
<td>Neocortical AChE deficit, total group: 32.2%</td>
<td>z in −1.5 to −2 Range Cognitive Subgroup (n = 11)</td>
<td>6 (85.7)</td>
<td>8 (72.7)</td>
</tr>
<tr>
<td>PPN-thalamic AChE deficit, total group: 16.8%</td>
<td>z &gt; −0.5 Cognitive Subgroup (n = 88)</td>
<td>2 (28.6)</td>
<td>2 (18.2)</td>
</tr>
</tbody>
</table>

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² Significant variable after Holm-Bonferroni correction.

* Statistical significances for the distribution differences for each of the neurochemical deficits between the 5 cognitive subgroups.

The χ² testing was performed for comparison of proportions of innervation deficits between groups. In addition to the analysis of independent effects of cholinergic and nigrostriatal dopaminergic degenerations in PD to predict global and cognitive domain z scores, we also performed a post hoc multiple regression analysis of possible interaction effects between the caudate nucleus dopaminergic and cortical cholinergic denervation using absolute vesicular monoamine transporter type 2 and AChE innervation variables. Duration of disease was used as a coregressor. All analyses were performed using SAS version 9.2 (SAS Institute). The Holm-Bonferroni correction for multiple testing was performed for each main analysis.
The combined presence of caudate nucleus dopaminergic and neocortical cholinergic deficits increased from 14.8% to 71.4% across the spectrum of cognitive impairment (total model, $\chi^2 = 37.8; P = .002$).

**Post Hoc Analysis: Interactive Effects Between Caudate Nucleus Dopaminergic and Cortical Cholinergic Degenerations**

Because the vast majority (87.0%) of patients with neocortical cholinergic deficits also had caudate nucleus dopaminergic deficits (Table 2), we investigated possible interactive effects between these 2 degenerations and cognitive functions. Table 3 shows the findings of the multiple regression analysis performed with cognitive z scores as outcome parameters and absolute cortical $[^{11}C]$PMP AChE activity levels and caudate nucleus $[^{11}C]$DTBZ vesicular monoamine transporter type 2 distribution volume ratios and their interaction term as regressors. The total model was significant for the global composite z score ($F = 7.51; P < .001$) and showed not only independent cognitive predictions for caudate nucleus dopaminergic ($F = 7.25; P = .008$) and cortical cholinergic ($F = 7.50; P = .007$).

**Figure. Frequency of Neocortical Cholinergic Deficits Across the Cognitive Spectrum**

The relative frequencies of significant neocortical cholinergic deficits in consecutive 0.5 z score ranges of cognitive impairment based on global cognitive z scores ranging from no or minimal ($z > -0.5$) to more severe cognitive impairment ($z \leq -2$) in a cohort of predominantly nondemented patients with Parkinson disease ($n = 143$).

**Table 2. Frequency Listing of the 4 Different Combinations of Presence or Absence of Significant Caudate Nucleus Dopaminergic and Neocortical Cholinergic Deficits in Cognitive Severity Subgroups**

<table>
<thead>
<tr>
<th>Combination of Absence or Presence of Neurochemical Deficits</th>
<th>z ≤ −2 Cognitive Subgroup (n = 7)</th>
<th>z in −1.5 to −2 Range Cognitive Subgroup (n = 11)</th>
<th>z in −1.0 to −1.5 Range Cognitive Subgroup (n = 10)</th>
<th>z in −0.5 to −1.0 Range Cognitive Subgroup (n = 26)</th>
<th>Statistical Significance, $\chi^2$</th>
<th>P Value</th>
</tr>
</thead>
<tbody>
<tr>
<td>Abnormal caudate nucleus VMAT2 and abnormal neocortical AChE activity, 28.2%</td>
<td>5 (71.4)</td>
<td>8 (72.7)</td>
<td>3 (30)</td>
<td>5 (19.2)</td>
<td>13 (14.8)</td>
<td>37.8 .002</td>
</tr>
<tr>
<td>Abnormal caudate nucleus VMAT2 and normal neocortical AChE activity, 42.3%</td>
<td>0 (0)</td>
<td>3 (27.3)</td>
<td>2 (20)</td>
<td>14 (53.9)</td>
<td>33 (36.4)</td>
<td></td>
</tr>
<tr>
<td>Normal caudate nucleus VMAT2 activity and normal neocortical AChE activity, 4.2%</td>
<td>1 (14.3)</td>
<td>0 (0)</td>
<td>2 (20)</td>
<td>0 (0)</td>
<td>9 (10.2)</td>
<td></td>
</tr>
<tr>
<td>Normal caudate nucleus VMAT2 and normal neocortical AChE activity, 25.3%</td>
<td>1 (14.3)</td>
<td>0 (0)</td>
<td>3 (30)</td>
<td>7 (26.9)</td>
<td>34 (38.6)</td>
<td></td>
</tr>
</tbody>
</table>

Abbreviations: AChE, acetylcholinesterase; VMAT2, vesicular monoamine transporter type 2.

*Statistical significance for the distribution differences for the 4 possible combinations of presence or absence of the 2 neurochemical deficits between the 5 cognitive subgroups.

**Table 3. Results From Regression Analysis With Cognitive z Scores as Outcome Parameters and Neocortical Acetylcholinesterase Activity Levels, Caudate Nucleus Vesicular Monoamine Transporter Type 2 Distribution Volume Ratios, and Interaction Terms as Regressors**

<table>
<thead>
<tr>
<th>Outcome Measure</th>
<th>Cortical Acetylcholinesterase Group Effect, F</th>
<th>P Value</th>
<th>Caudate Nucleus Vesicular Monoamine Transporter Type 2 Distribution Volume Ratio Covariate Effect, F</th>
<th>P Value</th>
<th>Interaction Term, F</th>
<th>P Value</th>
<th>Total Model, F</th>
<th>P Value</th>
</tr>
</thead>
<tbody>
<tr>
<td>Global composite z score</td>
<td>7.50 .007</td>
<td>7.25 .008</td>
<td>5.40 .022</td>
<td>7.51 &lt;.001*</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Cognitive subdomains</td>
<td>Verbal learning z score</td>
<td>4.49 .04</td>
<td>4.30 .04</td>
<td>2.98 .087</td>
<td>6.0 .002*</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Executive functions z score</td>
<td>8.98 .003</td>
<td>9.18 .0029</td>
<td>7.03 .009</td>
<td>7.38 &lt;.001*</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Visuospatial function z score</td>
<td>0.73 .40</td>
<td>0.43 .51</td>
<td>0.37 .55</td>
<td>0.81 .53</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Attention z score</td>
<td>5.88 .02</td>
<td>6.28 .013</td>
<td>4.63 .034</td>
<td>6.06 .002*</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
</tbody>
</table>

*Significant after Holm-Bonferroni correction.
Discussion

Our findings show that substantial caudate nucleus dopaminergic denervation is common even in patients with PD with minimal or no cognitive impairment. In contrast, cortical cholinergic degeneration is significantly less common in patients with PD with minimal cognitive impairment and becomes very frequent in patients with PD with greatest cognitive deficits. These data indicate that cortical cholinergic deficits are likely important contributors to the emergence of dementia in PD. Consequently, cortical cholinergic afferent degeneration is probably a major process associated with progressive cognitive decline across the spectrum of cognitive impairment in PD. These results agree with prior cholinergic in vivo imaging and postmortem studies showing that cortical cholinergic denervation can occur early in some patients with PD but that progressive and more extensive cortical cholinergic denervation is characteristic of PD dementia.3,4,22-26

Although dopaminergic denervation of the caudate nucleus and frontolimbic system alone may not be sufficient for the development of marked cognitive impairments in PD,3,24 cortical cholinergic denervation occurs mainly in patients with significant caudate nucleus dopaminergic denervation. Our post hoc regression analysis showed significant interaction effects with combined effects of these 2 degenerative processes implicating their contributions to worsening cognitive impairment in PD, indicating that caudate nucleus dopaminergic and cortical cholinergic deficits may contribute to cognitive impairment in PD, both additively and multiplicatively. A recent rat study evaluated cognitive effects of either selective dopaminergic denervation of the rodent homologue of the caudate nucleus and/or lesions of frontal cortical cholinergic afferents.27 Dual forebrain dopaminergic and cholinergic lesions induced greater deficits on a sustained attention test compared with effects of isolated lesions of either system alone.27 Isolated dopaminergic lesions actually enhanced attentional performance. As attentional function in this behavioral paradigm is mediated by cortical cholinergic afferents, this result suggested that caudate homologue dopaminergic denervation induced compensatory overactivity of cortical cholinergic afferents. These observations suggested that dual dopaminergic-cholinergic lesions resulted in the loss of attentional cognitive resources that compensated for cognitive deficits resulting from striatal dysfunction.26

This model is supported by our data. We found that significant caudate nucleus dopaminergic denervation was present in about two-thirds of patients with essentially normal global cognition (global cognitive composite z score greater than −0.5) but a high proportion of patients with markedly impaired cognition (global cognitive z score of less than −2.0) had dual dopaminergic-cholinergic deficits. Also consistent with the model of enhanced cholinergically mediated attentional function compensating for caudate dopaminergic deficits were the results of our regression analysis, demonstrating that attentional and executive functions, which are strongly influenced by cortical mechanisms but not visuospatial function deficits, were strongly related to combined dopaminergic-cholinergic deficits. Relatively isolated impairment of a single neurotransmission system in PD (ie, nigrostriatal denervation) may not lead to the development of significant cognitive impairments because of adaptive plasticity in other intact brain systems.28-30

Attempts to reconcile the existence of multisystem degenerations and cognitive impairments in PD have been dominated by models suggesting direct correlations between single neurochemical system changes and distinct cognitive deficits.31 These models are undoubtedly too simplistic. Our findings support more complex pathophysiological models of interacting dopaminergic and cholinergic degenerative changes producing cognitive dysfunctions in PD.28,31

Our results are relevant also to evaluating the dual syndrome hypothesis of Kehagia et al,2 which posits early executive deficits secondary to frontostriatal system dysfunction driven by caudate dopaminergic denervation and, later, more global deficits owing to multiple pathologies, including cholinergic system degeneration, that affect more posterior cortices. However, our results are more consistent with cholinergic deficits exacerbating frontostriatal dysfunction owing to the loss of compensatory frontal cortical attentional functions, perhaps resulting in additional dysfunctional corticostriate signaling.28 This interpretation is also more consistent with recent findings that cholinergic afferents are relatively enriched in frontal cortices.32

One limitation of our study was its cross-sectional design, which lacked longitudinal information about the rate of conversion to dementia and lack of inclusion of patients with more severe dementia (in whom commonly prescribed use of cholinesterase inhibitor drug therapy precluded participation in the present study). Another limitation was that our monoaminergic PET ligand was not suited to assess extrastriatal mesofrontal or limbic changes. The lack of visuospatial function findings in our study may be related to differences in the cognitive test battery between our study and other studies.2 Our study included more men than women. It is possible that sex differences in PD incidence and age at onset may be owing to the possible neuroprotective effects of estrogen in PD.33 However, while it is a possibility, the difference in sex would not be expected to affect a relative difference of degree of denervation between the dopaminergic vs the cholinergic systems in individuals who already have PD. Finally, we did not assess other α-synucleinopathy–associated processes, such as degeneration of noradrenergic projections or cortical α-synucleinopathy that likely contribute to impaired cognition in PD.34
Conclusions

The heterogeneous basis of cognitive deficits in PD suggests multiple complementary approaches to symptomatic treatment. Our data provide a good rationale for improved cholinergic augmentation therapy in patients with PD with cognitive symptoms. Caudate nucleus and cortical cholinergic deficits contribute to cognitive impairment in PD not only in an additive but also in a synergistic fashion.

REFERENCES


