New Magnetic Resonance Imaging Biomarkers Advance the Characterization of Parkinson Disease

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Abstract
The pathophysiology of idiopathic Parkinson disease (PD) is traditionally characterized as substantia nigra degeneration, but careful examination of the widespread neuropathologic changes suggests individual differences in neuronal vulnerability. A major limitation to studies of disease progression in PD has been that conventional magnetic resonance imaging (MRI) techniques provide relatively poor contrast for the structures that are affected by the disease, and thus are not typically used in experimental or clinical studies. Here, we review the current state of structural MRI as applied to the analysis of the PD brain. We also describe a new multispectral MRI method that provides improved contrast for the substantia nigra and basal forebrain, which we recently used to show that these structures display different trajectories of volume loss early in the disease.

Keywords
Parkinson’s disease, MRI, substantia nigra, basal forebrain, neurodegeneration, biomarkers

Parkinson disease (PD) is a devastating neurodegenerative disorder characterized by its cardinal motor symptoms: resting tremor, muscular rigidity, bradykinesia, postural instability, and gait abnormality. PD currently affects 1–2% of individuals over age 65, totaling five million people worldwide. During the next 20 years, the incidence of PD is projected to double, making research on its causes and treatments timelier and more relevant to global public health than ever before. A major impediment to such research is the paucity of safe, fast, and effective brain imaging methods for visualizing the structures affected by PD. Because conventional structural magnetic resonance imaging (MRI) techniques cannot visualize the brain changes that are at the core of this disease, MRI-based biomarkers for diagnosis and tracking disease progression do not currently exist.

The cardinal motor features of PD are typically attributed to a loss of nigrostriatal dopaminergic neurons in the substantia nigra pars compacta (SNpc), which is accompanied by the aggregation of Lewy bodies and neurites in this structure. While denervation of dopaminergic nigrostriatal projections may explain the primary motor symptoms of PD, as shown by the dramatic motor improvement associated with dopamine replacement therapy, abnormalities beyond the SNpc likely underlie the serious and potentially debilitating non-motor features, including cognitive and memory impairments and progression to dementia. Notably, degeneration of the cholinergic basal forebrain (BF) and noradrenergic locus coeruleus in PD probably contribute to non-motor deficits. Although deterioration in the BF and LC is most often associated with late-stage PD with dementia, subtle changes in earlier stages could result in poor performance on tests of memory and attention. Research on these non-motor aspects of the disease has been hindered by a lack of sensitive MRI biomarkers for the affected structures. This article reviews recent progress in developing new MRI-based biomarkers to visualize and characterize abnormalities in some of the brain structures affected by PD.

Imaging the Substantia Nigra
The substantia nigra comprises two structurally and functionally segregated regions: the SNpc, which projects mainly to the striatum and basal ganglia, and the substantia nigra pars reticulata (SNr), which sends its primary efferent projections to the thalamus and superior colliculus. In PD, neuronal loss in the SNpc is prevalent in the caudal and mediolateral part and more limited in mediorostral areas. This loss of SNpc neurons results in a marked depletion of dopamine in the striatum, and to a lesser extent in other basal ganglia nuclei. The pattern of dopamine loss in the striatum parallels the lateral to medial gradient of cell loss in the SNpc, with cells projecting to the putamen showing signs of atrophy first, followed by those that project to the caudate nucleus and nucleus accumbens. Functionally segregated circuits link the basal ganglia and cortex in a topographical manner, with dense reciprocal fronto-striatal connections, which are known to support high-order cognitive functions. Because abnormalities in any part of the complex basal ganglia-thalamocortical circuitry could have significant downstream consequences, PD may be considered a network disease. Significant progress has been made using MRI to accurately segment the structures of the basal ganglia, but few tools exist for measuring the size and structure of the SNpc.

One hindrance to the development of effective morphometric tools is that the borders of the SNpc are nearly impossible to visualize on conventional T1-weighted MRI. As a result, numerous attempts have...
been made to develop new sequences that would provide indices of nigral degeneration in PD. \textsuperscript{24} Results from these studies, however, are wrought with contradictions. Early attempts at visualizing the SNpc capitalized on the relative distribution of iron in the midbrain, which causes magnetic susceptibility artifacts and signal loss in T2-weighted images. \textsuperscript{29} The SNr has relatively high levels of iron, and thus appears as a hypointense region, whereas the dopaminergic SNpc, which contains neuromelanin, appears as a hyperintense region between the SNr and red nucleus on axial images. \textsuperscript{26} Although most studies reported signal loss or reduced size of the SNpc in PD patients compared to controls, \textsuperscript{31–35} some failed to find disease-related changes, \textsuperscript{39,40} and others pointed out potential confounds in prior studies. \textsuperscript{26} Limitations in visualizing the SNpc in T2-weighted images were that the anatomical location of the SNpc appeared to be inconsistent with histologic reports, and researchers were not able to reliably differentiate SNpc from SNr. \textsuperscript{30} A subsequent study used proton density-weighted MRI in combination with short inversion-time recovery images to more accurately distinguish the SNpc and SNr, but did not find a significant decrease in SNpc size in PD patients. \textsuperscript{20}

Newer methods, such as the use of MR sequences sensitive to neuromelanin, \textsuperscript{46} evaluation of T2 relaxation times, \textsuperscript{41} and segmented inversion recovery ratio imaging \textsuperscript{22–24} have achieved greater success in differentiating SNr and SNpc and have documented changes in the SNpc in PD. Further, T2*- and diffusion-weighted imaging methods have been found to be sensitive to disease-related changes in the SNpc, likely as a result of differences in MR inhomogeneities related to the relatively high iron content of this structure. \textsuperscript{45,46} An emerging method, connectivity-based segmentation of the SN using diffusion tensor imaging, may prove useful for delineating SNpc in PD. \textsuperscript{47} We recently described a new multispectral MRI method for visualizing the SNpc. \textsuperscript{48} Our multispectral sequences included multiecho MPRAGE with T1-weighting, multiecho Fast Low-Angle Shot (FLASH) with proton density weighting, 3D T2 SPACE turbo spin echo, and 3D T2 SPACE fluid-attenuated inversion recovery (FLAIR) turbo spin echo. For anatomical analyses of cortical structures, we showed that high-bandwidth T1-weighted multiecho MPRAGE data were superior to conventional T1-weighted images. \textsuperscript{26,49} Further, multiecho sequences were less prone to distortion and had a higher contrast-to-noise ratio for subcortical structures. These sequences were bandwidth-matched at 698 Hz/pixel—a property that was critical for facilitating coregistration across scans without distortion corrections. Because we could perform precise spatial registration of scans that had different contrasts, we could generate multiple weighted averages of the sequences, each with a unique contrast tailored to a specific set of structures. Thus, multispectral data yield a broad range of contrasts that allow for enhanced anatomical analysis and provide valuable new data about the subcortical structures implicated in PD.

We generated weighted averages of scans with different contrasts, emphasizing the contribution of proton density- and T2-weighted images, with a lesser, but important, contribution from T1-weighted and T2-FLAIR images. This method allowed reliable delineation the SNpc in a relatively large sample of PD patients and controls. The fact that the anatomical location of the SNpc in our images corresponded well to those described in the most accurate MRI studies performed to date, \textsuperscript{20,40,43} increased our confidence in the utility of this method for distinguishing SNpc from SNr. As a result, we were able to detect a significant decrease in the volume of the SNpc in the earliest stage of the disease (see Figure 1).

**Imaging the Basal Forebrain**

The BF is a collection of cholinergic nuclei that contains the diagonal band of Broca, medial septum, and nucleus basalis of Meynert. \textsuperscript{31} These nuclei constitute the primary source of cholinergic innervation of the entire cerebral cortex \textsuperscript{12–14} and are essential for a host of cognitive processes, including attention and long-term memory. \textsuperscript{55–58} Degeneration of the BF is often considered a hallmark of Alzheimer’s disease (AD) pathology, \textsuperscript{57,59–61} but some studies suggest that cell loss and cholinergic dysfunction in PD is comparable or even greater. \textsuperscript{61,62}

Direct confirmation of cholinergic degeneration in PD comes from neuropathologic studies that uncovered a pronounced loss of cholinergic neurons in the BF of patients with PD. \textsuperscript{15,51,61,63,64} In addition, another post mortem investigation documented decreases in the biochemical markers of cholinergic function, including choline acetyltransferase (ChAT) and acetylcholinesterase (AChE). \textsuperscript{65} Researchers found decreased ChAT activity in non-demented PD patients and an even greater drop in those with dementia. Several reports linked non-motor cognitive impairments to cholinergic dysfunction. \textsuperscript{66,67} PET experiments provided complementary in vivo confirmation of a loss of cholinergic function in PD, in particular a marked reduction of AChE activity in cortical regions. \textsuperscript{68–70} Based on known patterns of cholinergic projections, \textsuperscript{12–14} it appears likely that this decrease in cortical AChE activity is caused by a loss of cholinergic neurons in the BF. Although AChE dysfunction appears relatively early in PD, \textsuperscript{46,71} the magnitude of the disruption is greater and more widespread in PD patients with dementia than in those without. \textsuperscript{69,70} Other PET studies confirmed the presence of altered cholinergic neurotransmission in patients with mild PD. \textsuperscript{65,69,72} What was lacking until recently was a thorough MRI-based in vivo examination of the morphology of the BF early in the disease. \textsuperscript{73,74}

MRI studies of the BF have generally relied on T2-weighted images. In contrast to studies of the SNpc, relatively few studies have attempted to use...
MRI to measure disease-related changes in the BF of PD patients. Studies of patients with AD and other forms of dementia have resolved the BF (i.e., the substantia innominata) on T2-weighted images.75,76 Using similar methods, one study demonstrated reduced thickness of this structure in demented PD patients in the later stages of the disease.77 The volume of the BF has also been measured using T1-weighted images collected at 3T, in which the borders of the BF are more visible than in lower field strength images.78 This method revealed a significant decrease in the volume of the BF between non-demented PD patients and controls, with further volume loss in demented patients.79 Few studies, however, have described the morphology of this structure in early-stage, non-demented PD patients.

In addition to using our new multispectral MRI methods to measure the SNpc,48 we found that our T2-FLAIR images provided superior contrast for the BF than that achieved in previous MRI studies using standard T2-weighted images. We were, therefore, able to examine the morphology of this structure in the same set of PD patients and controls. In contrast to our finding of decreased SNpc volume in the earliest stages of the disease, BF volume loss occurred later in the disease, with a significant decrease apparent only in patients in Hoehn and Yahr Stages 2 and 3, but not in Stage 1 patients. This finding of greater BF volume loss at later stages of the disease is consistent with the results from previous PET studies, as well as with reports of a more dramatic degree of BF cell loss in PD patients with dementia, compared to non-demented patients.51

Comparison of Substantia Nigra Pars Compacta and Basal Forebrain Changes in Parkinson’s Disease

According to the influential Braak neuropathologic staging scheme, progression of neurodegenerative processes in PD (e.g., Lewy body deposition) begins in the enteric and peripheral nervous system, and then progresses anteriorly to the brainstem, midbrain, forebrain, and neocortex.81,82 While this account is based on a careful neuropathologic examination of a large sample of post mortem specimens, definitive confirmation of the hypothesis is lacking.83 Validation requires in vivo indices of the timing and progression of neuronal degeneration in these areas. A specific hypothesis that arises from this staging scheme is that pathologic changes in the SNpc should precede degeneration of the more anterior BF.83,84 Until recently, this hypothesis had not been tested, because the morphology of the SNpc and BF had been examined only in separate study groups using data collected at different times.

We applied our newly developed multispectral MRI techniques to test the hypothesis that degeneration of the SNpc precedes the BF. New multispectral structural MRI sequences allowed us to visualize and measure disease-related changes in both structures in a single sample of PD patients and controls. Consistent with this hypothesis, our analysis of MRI-derived volumetric data revealed a decrease in the volume of the SNpc in patients who were in early stages of the disease, but lower BF volumes only in patients with more advanced PD.85 This study provided the first direct in vivo support for the Braak neuropathologic staging scheme in PD.85,86 Our MRI data provide a complement to existing neuropathologic studies, which are based entirely on measures of α-synuclein inclusions and not on cell or volume loss, with uncertainty regarding the existence of a correspondence between α-synuclein pathology and loss of volume.86 Early pathologic insults to non-dopaminergic nuclei underlie deficits in attention and cognitive control in the early stages of PD. The identification of clusters of patients with distinct patterns of cognitive impairment raises the possibility that a subgroup of idiopathic PD patients may be at greater risk for developing memory impairments and dementia, which may stem from exacerbated BF degeneration, and more extensive disruption of cholinergic innervation of the neocortex.87,88 Evidence of such heterogeneity stems from the observation that some advanced non-demented PD patients actually showed higher levels of AChE activity than a sample of drug-naïve patients in the earliest stages of the disease.89 Pharmaceutical augmentation of cholinergic activity led to a significantly reduced frequency of failing in some PD patients.90 Further, PD patients with postural and gait disturbances were at greater risk for developing dementia.91

Parallels Between Parkinson’s Disease and Alzheimer’s Disease in Patterns of Basal Forebrain Degeneration

The fact that cholinergic degeneration in PD worsens with disease progression and is exacerbated in PD patients with dementia51,69,70,92 suggests parallels between PD and AD. While disruption of cholinergic function appears to be linked to the development of cognitive impairments and dementia, explicit verification of a shared pathogenic mechanism between PD and AD remains equivocal. In PD, neuronal loss or Lewy body deposition in the BF can occur in the absence of the hallmark neuropathologic features of AD—amyloid plaques and neurofibrillary tangles.93 Other studies in PD patients have failed to find a significant correlation between level of cognitive impairment and degree of cell loss96 or Lewy body burden97 in the BF, leading to the hypothesis that the degree of cholinergic degeneration must reach a critical threshold before the symptoms of dementia emerge.98 Thus, while some have proposed a common mechanism underlying BF degeneration in AD and PD with dementia, direct support for this hypothesis is lacking. An alternative proposal is that BF degeneration is primary in PD, whereas the cholinergic cell loss in AD is secondary to cortical pathology, which leads to depleted retrograde transport of critical growth factors to BF neurons.99

Whether a bona fide pathologic parallel exists between PD and AD, the disruption of cholinergic function in PD has real and important clinical implications that are not limited to dementia. Cholinergic dysfunction has been linked to a number of non-motor symptoms, including decreased performance on tests of working memory,70 set-shifting,99,100 and free recall.101 In addition, administration of anticholinergic drugs led to the development of executive102 and memory103 impairments in PD patients who did not previously show these deficits. Some of these symptoms were partially ameliorated by treatment with central cholinesterase inhibitors,102–104 and indeed, cholinesterase inhibitors have become a staple in the treatment of PD with dementia.105,106

Conclusions

Recent advances in structural MRI technology have laid the groundwork to the development of new biomarkers to visualize and characterize abnormalities that are at the core of PD. As these methods become refined and are applied to large samples of PD patients, they will enable scientists and physicians to derive and track disease progression in subgroups of patients, thereby paving the way for specialized clinical treatments based on a detailed profile of brain changes.
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Movement Disorders  Parkinson’s Disease

101. Dubois B, Daran F, Pillon B, et al., Cholinergic-dependent cognitive deficits in Parkinson’s disease, Ann Neurol,