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Cholinergic Dysfunction in Parkinson's Disease

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Abstract

There is increasing interest in the clinical effects of cholinergic basal forebrain and tegmental pedunculo-pontine complex (PPN) projection degeneration in Parkinson's disease (PD). Recent evidence supports an expanded role beyond cognitive impairment, including effects on olfaction, mood, REM sleep behavior disorder, and motor functions. Cholinergic denervation is variable in PD without dementia and may contribute to clinical symptom heterogeneity. Early *in vivo* imaging evidence that impaired cholinergic integrity of the PPN associates with frequent falling in PD is now confirmed by human post-mortem evidence. Brainstem cholinergic lesioning studies in primates confirm the role of the PPN in mobility impairment. Degeneration of basal forebrain cholinergic projections correlates with decreased walking speed. Cumulatively, these findings provide evidence for a new paradigm to explain dopamine-resistant features of mobility impairments in PD. Recognition of the increased clinical role of cholinergic system degeneration may motivate new research to expand indications for cholinergic therapy in PD.

Keywords

α -Synuclein; Acetylcholine; Acetylcholinesterase; Basal forebrain; Brainstem; Cognition; Dementia with Lewy bodies; Dopamine; Falls; Gait; Lewy bodies; Mood; Nucleus basalis of Meynert; Olfaction; Parkinson's disease; Pedunculo-pontine nucleus; Pontine tegmentum; Positron Emission Tomography (PET); Posture; Progressive supranuclear palsy; REM sleep behavior disorder; Single Photon Emission Computed Tomography (SPECT); Striatum; Thalamus; Cholinergic Dysfunction

Introduction

The traditional view of Parkinson's disease (PD) as a predominant single-system neurodegeneration characterized by nigrostriatal dopaminergic denervation secondary to

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Compliance with Ethics Guidelines

Conflict of Interest

Martijn L.T.M. Müller, PhD^{1*} & Nicolaas I. Bohnen declare that they have no conflict of interest.

Human and Animal Rights and Informed Consent

This article does not contain any studies with human or animal subjects performed by any of the authors.

neuronal Lewy body inclusions is waning [1]. Instead, PD is increasingly viewed as a multisystem neurodegeneration affecting multiple neurotransmission systems [2]. Of particular interest, and an increasing focus of research investigations, is degeneration of the cholinergic system in PD. In Alzheimer's disease (AD) research, degeneration of the cholinergic system has provided a successful direction for neurobiological and drug development research. However, it has been recognized that cholinergic denervation can occur also in PD and is often more severe than in AD [3]. In this article we will provide a brief overview of cholinergic mechanisms, neuropathological findings, and *in vivo* imaging markers, which has been reviewed more extensively previously [4]. The main goal of the review is to provide an update on the latest research findings regarding the clinical sequelae of cholinergic dysfunction in PD, with an emphasis on the role of the cholinergic system in posture and gait control in PD.

1. Cholinergic system projections and receptors

Acetylcholine is a ubiquitous neurotransmitter with widespread innervation in the cortex, subcortical structures, and the cerebellum. There are three major sources of cholinergic projections in the brain. Basal forebrain (BF) neurons, including the nucleus basalis of Meynert (nbM), provide the cholinergic projections to the cerebral cortex [5]. The pedunculopontine nucleus-laterodorsal tegmental complex (henceforward referred to as the PPN), a brainstem center, provides cholinergic inputs primarily to the thalamus, but also has connections to the cerebellum, several brainstem nuclei, some striatal fibers, and the spinal cord [6]. In addition, small populations of intrinsic cholinergic neurons are present in the hippocampus, striatum (cholinergic interneurons), cortex, the medial habenula, parts of the reticular formation, and cerebellum [7–10].

Two important subtypes of cholinergic receptors are the nicotinic (nAChR) and muscarinic (mAChR) receptors. The majority of nAChRs in the brain consist of the $\alpha 4\beta 2$ subtype [11]. Nicotinic receptor binding is high in the thalamus, striatum, and substantia nigra pars compacta, and is also present in the cerebellar cortex and dentate nucleus [12]. Muscarinic receptors also exist as several subtypes. High concentrations of mAChRs can be found in the neocortex, hippocampus, olfactory tubercle, and amygdala. The highest concentration of muscarinic receptors is found in the striatum [13, 14].

2. Cholinergic denervation in PD, PDD/DLB, and PSP

Neuropathological research on degeneration of the cholinergic system has had a traditional focus on AD. Findings show that there is no generalized cholinergic lesion in AD but rather selective cortical cholinergic denervation [15, 16]. Studies indicate that cortical cholinergic deficits are not severe in mild AD and become prominent only in late-stage disease [17–20]. In advanced AD, loss of cholinergic innervation in the cerebral cortex is widespread, with the most severe losses in the temporal lobes [21, 22]. These cortical cholinergic losses in AD have primarily been associated with cognitive impairment and dementia [23].

Several decades of neuropathology research has generated considerable evidence for altered cholinergic neurotransmission in PD, PD with dementia (PDD) and dementia with Lewy bodies (DLB). The pathological hallmark of PD is deposition of Lewy body pathology in the

brainstem. The subsequent loss of midbrain dopaminergic neurons of the substantia nigra, pars compacta, results in striatal dopaminergic denervation. An interesting fact is that Frederic Lewy first identified the eponymous Lewy body, of which α -synuclein fibrils are the structural component, not in the substantia nigra but in the nbM [24]. In the Braak temporal staging proposal of PD pathology, Lewy bodies and neuronal loss in the substantia nigra occurs concurrently with accumulation of α -synuclein deposition in cholinergic neurons of the BF [2]. This indicates that cholinergic denervation occurs early in PD. Significant loss of nbM cholinergic neurons has been reported extensively in PD brains, especially in the presence of dementia [25–29]. The extent of nbM cholinergic loss is profound; greater neuronal loss of nbM neurons has been reported in PD compared to AD [30]. Unlike AD, where PPN cholinergic neurons are generally preserved [16, 31], neuropathological studies of PD have reported that about 50% of the large cholinergic neurons of the lateral part of the PPN, pars compacta, degenerate [31–34].

A clinical distinction is made between PD patients who develop dementia after the onset of parkinsonism and DLB patients who develop dementia ahead of or concurrently with parkinsonism [35]. Although pathological findings of Lewy body pathology and cholinergic degeneration are comparable in DLB to PDD, amyloid plaque deposition is found in a majority of DLB compared to a minority of PDD cases [36–40].

Progressive supranuclear palsy (PSP) is an atypical parkinsonian syndrome associated with early severe impairment of gait and balance [41]. Cholinergic positron emission tomography (PET) studies in PSP subjects indicate a greater loss of thalamic acetylcholinesterase (AChE) activity than in patients with PD, reflecting significant loss of brainstem cholinergic pedunculopontine neurons [31, 33]. However, cortical cholinergic innervation is relatively spared in PSP [42, 43].

3. In vivo PET and SPECT cholinergic system findings in PD and PDD/DLB

PET and single photon emission computed tomography (SPECT) studies allow *in vivo* visualization and quantification of the brain cholinergic system. Targets for PET and SPECT radiotracers are constituents of acetylcholine synthesis, storage, and recycling. The two most widely used PET tracers are the acetylcholine analogues [^{11}C]methyl-4-piperidyl acetate ([^{11}C]MP4A) and [^{11}C]methyl-4-piperidiny propionate ([^{11}C]PMP). These tracers are metabolized and then trapped by AChE, and as such reflect AChE distribution. AChE is a reliable marker of brain cholinergic pathways and its regional distribution has a good correspondence with that of choline transferase activity [44]. Radiotracer distribution measured with AChE PET imaging shows highest activity in the basal ganglia and the BF nuclei, intermediate in the cerebellum, and lowest activity in the cortex, similar to biodistribution findings in post-mortem brains [45]. AChE PET imaging assesses cholinergic terminal integrity with cortical uptake reflecting largely BF neuron integrity and thalamic uptake principally reflecting PPN complex integrity [4].

The cholinergic denervation revealed by molecular neuroimaging in PD is generally in agreement with that revealed by histopathology. *In vivo* cholinergic PET studies have identified cholinergic deficits in PD without dementia and PDD patients (Figure 1) [42, 46, 47]. PD patients without dementia show less severe but significant reductions in cortical

AChE activity most prominent in the medial secondary occipital cortex [3, 48]. Furthermore, reductions in cortical AChE levels are greater and more extensive especially in PDD patients compared to AD patients with similar severity of dementia [3]. Two studies found that the degree of cholinergic loss between PDD and DLB is comparable indicating that the degree of BF cholinergic neuron pathology is similar between these two disease entities [48, 49]. It remains unclear, however, whether the degree of cholinergic denervation is similar between PDD and DLB at onset of these diseases. A recent *in vivo* AChE PET study suggested a variable degree of cholinergic dysfunction in early DLB, however, this was not compared to PD patients [50]. Greater cortical cholinergic loss in DLB at disease onset may potentially explain the earlier manifestation of cognitive and neurobehavioral impairments compared to PD(D).

Studies also show a widespread loss of cholinergic nicotinic and muscarinic receptors in PD. For example, a post-mortem *in vitro* study using a nAChR ligand demonstrated nAChR loss in the striatum that paralleled the loss of nigrostriatal dopaminergic markers [51]. *In vivo* imaging studies of PD subjects without dementia have also reported reductions of $\alpha 4\beta 2$ nAChR binding in the striatum, substantia nigra, and cortex [52, 53]. Meyer *et al.* reported more widespread loss of $\alpha 4\beta 2$ nAChR binding in PD without dementia, including the midbrain and cerebellum [54]. A muscarinic receptor PET study demonstrated increased mAChR binding in the frontal cortex in PD patients without dementia [55]. This finding may reflect either greater receptor availability or an adaptive up-regulation in response to denervation of the BF and loss of ascending cholinergic input to the frontal cortex.

4. Heterogeneity of cholinergic denervation in PD

The degree of cholinergic denervation appears to be variable in PD. *In vivo* imaging studies have shown cholinergic projection losses that vary from 5% to 25% in PD subjects both with and without dementia [3, 42, 46, 48]. We recently examined heterogeneity of cortical and thalamic cholinergic denervation in PD [56]. We determined *in vivo* cortical and thalamic AChE activity with PET imaging in PD patients without dementia and in healthy non-PD subjects. AChE activity of the cortex and thalamus was compared to cholinergic innervation of these regions in the healthy non-PD control subjects and dichotomized PD patients as within-normal-range or below-normal-range based on a 5th percentile cutoff from normal. The results of our study found that neocortical and thalamic AChE activity were within-normal-range for 65 out of 101 PD subjects. The remainder had combined neocortical and thalamic, isolated neocortical, or isolated thalamic denervation based on the 5th percentile cutoff from normal. The variable pattern of BF and brainstem cholinergic denervation is predicted to reflect clinical phenotypic variations in these patients.

5. Clinical correlates of cholinergic denervation in Parkinson's disease

There is accumulating evidence that cholinergic system degeneration not only contributes to cognitive but also to other non-motor features and motor impairments in PD. The effect of selected cortical and thalamic cholinergic denervation on cognition, olfaction, mood, REM sleep behavior disorder, and balance and gait functions will be reviewed below.

5.1 Cognition—Impairments in cognition and dementia are frequent findings in PD patients. Approximately 75% of the PD patients who survive for more than 10 years will develop dementia [57]. In PD patients without dementia, executive dysfunction can often be noted manifesting as impaired cognitive flexibility, decision making, and working memory early in disease [58]. This executive dysfunction may be attributable to striatal dopaminergic denervation. Indeed, an *in vivo* dopamine PET imaging study showed that dopamine plays a significant role in working memory processes in PD [59]. However, while dopaminergic therapy in PD patients selectively improves some functions, over time cognitive impairment does not significantly respond to dopaminergic agents [60].

Neuropathological studies have shown that cognitive impairment in PD is associated with BF cholinergic loss. For example, reductions of AChE activity in frontal cortices are greater in PDD compared to PD patients without dementia [61]. Furthermore, the degree of cognitive impairment correlates with reduced choline transferase activity in the prefrontal cortex in the absence of specific AD-type pathology [62]. Reduced number of D1 dopamine receptors in the caudate nucleus also correlated with cognitive impairment in these patients [62].

Findings from our AChE PET studies show that cholinergic denervation associates with poorer performance on cognitive tasks [63]. Moreover, our recent *in vivo* AChE PET finding shows that cognitive performance is more severe in the cortical below-normal-range compared to the within-normal range cholinergic innervation PD sub-group (see also section 4). In addition, we found an independent effect of nigrostriatal dopaminergic denervation on cognition. These *in vivo* and the neuropathological findings support the “dual syndrome” hypothesis of cognitive impairment in PD by Kehagia *et al.* [64]. This hypothesis predicts independent contributions of both dopaminergic denervation on fronto-striatal cognitive, including executive, impairments and cholinergic denervation on visuospatial and other attentional impairments in PD. Williams-Gray and colleagues reported that more posterior cortically-based cognitive defects evolved over time into dementia, whereas, frontostriatal executive deficits were not associated with subsequent dementia risk *per se* [65]. These data agree with dopaminergic PET studies showing that dopaminergic denervation of the caudate nucleus and fronto-limbic system alone is not sufficient for PD dementia [46, 49]. However, dual-radiotracer dopaminergic and cholinergic PET imaging studies agree with the post-mortem evidence that primary BF cholinergic system degeneration significantly increases with the appearance of dementia [46, 49].

Reductions in nAChRs also parallel cognitive dysfunction in PD. For example, neuropathological studies have shown more severe reductions in cortical nAChRs in PD subjects that are demented [66, 67]. An *in vivo* nAChR PET imaging study confirmed these observations [54]. Overall, these results show that dopaminergic denervation results in impairment in selected cognitive domains, especially early in disease. However, convergent *in vivo* imaging and post-mortem evidence indicate that progressive cognitive decline resulting in dementia in PD is associated with BF cholinergic system degeneration [68].

5.2 Mood—Depressive mood, major depression, and apathy are common in PD [69]. Although degenerations of the monoaminergic systems have been linked to mood changes,

there is also evidence that this may be related to cholinergic system dysfunction in PD, at least in the presence of dementia. For example, the severity of cortical cholinergic denervation is associated with the presence of depressive symptoms and apathy in PD and PDD [70]. Depression in PD appears to be associated with cognitive deficits, which suggests a common mechanism [71]. This hypothesis is substantiated by the observation that depression is a risk factor for dementia in PD [72, 73]. Meyer *et al.* reported *in vivo* reductions of $\alpha 4\beta 2$ nAChRs in PD that correlated with both increased severity of depressive symptoms and severity of cognitive symptoms [54]. Depression in PD, at least in part, may be associated with the cholinergic system [74].

5.3 Olfaction—Impairment of olfactory function is highly prevalent in PD with reports indicating up to 95% of patients affected [75]. The cholinergic system modulates several aspects of central olfactory functions [76]. In our cholinergic AChE PET studies we have shown a robust cholinergic component to olfactory dysfunction in PD [77]. For example, we found that worse performance on the University of Pennsylvania Smell Identification Test (UPSIT) was associated with lower limbic and cortical AChE activity [78]. Although decreased striatal and hippocampal dopaminergic activity was also associated with lower UPSIT scores [79], limbic cholinergic denervation proved to be the most significant predictor of hyposmia in a multivariate analysis. In agreement with the hypothesized common cholinergic mechanism underlying cognition and odor identification, we found that odor identification scores correlated positively with scores on cognitive measures of episodic verbal learning [78]. Olfactory dysfunction in PD may be the most marked in subjects at risk of progressive cognitive decline, and may reflect the transition of PD toward a stage with more heterogeneous multisystem neurodegenerations [76].

5.4 REM sleep behavior disorder—Rapid eye movement (REM) sleep behavior disorder (RBD) is common in PD and is characterized by loss of normal atonia during REM sleep, resulting in abnormal motor manifestations (“acting out of dreams”). A common cholinergic mechanism may exist between RBD and the subsequent increased risk of dementia in PD [80–82]. For example, we recently showed that RBD was associated with cholinergic denervation. In our study, 33.8% of 80 non-demented PD subjects indicated a history of RBD symptoms, as defined by response on the Mayo Sleep Questionnaire. Subjects underwent dual-tracer AChE and dopaminergic PET imaging. PD subjects with RBD symptoms exhibited decreased neocortical, limbic cortical, and thalamic cholinergic innervation compared to those without RBD symptoms [83]. As RBD is an antecedent marker of cognitive impairment and dementia in PD, these findings suggest cholinergic system degeneration may occur early in PD [84].

5.5 Postural Instability and Gait Disorder—Postural instability and gait disorder (PIGD) is a significant cause of disability, and responds poorly to dopaminergic replacement except in the early phase of the disease [85, 86]. Accumulating evidence indicates that degeneration of the cholinergic system is a major contributor to PIGD features in PD [43]. In particular, we will review both primate and human findings that provide evidence for the putative role of PPN complex and BF cholinergic degeneration in the etiology of PIGD features in PD.

5.5.1 PPN cholinergic projections and mobility in primates: A recent primate study by Karachi, Grabli, and colleagues provides strong support for a critical role of PPN cholinergic neuron loss in PD-related postural dysfunction [87]. In a series of experiments young and old rhesus monkeys were lesioned with the neurotoxin 1-methyl-4-phenyl-1,2,3,6-tetrahydropyridine (MPTP) to induce a parkinsonian syndrome. MPTP lesioning resulted in nigrostriatal dopaminergic losses that were similar between young and old monkeys; however, additional loss of PPN cholinergic neurons was only observed in the older monkeys. Although both young and old monkeys manifested clinical parkinsonism, postural deficits were exclusively observed in the older monkeys. These results indicate that PPN cholinergic loss superimposed on nigrostriatal losses appears to be a critical factor for the emergence of postural problems in these parkinsonian monkeys.

Karachi *et al.* [87] continued to test the hypothesis that isolated PPN cholinergic loss (i.e. without dopaminergic injury) is sufficient to induce a postural and gait deficit in the primates. For this purpose, the investigators performed selective lesions of cholinergic PPN neurons using a specific neurotoxin in a different group of monkeys. The significant finding from this experiment was that isolated cholinergic PPN neuron lesioning, in the absence of nigrostriatal dopaminergic lesioning, led to prominent deficits in gait and posture in these monkeys. These deficits included a decrease in the angle of the knee, increased back curvature, and abnormal tail position (axial rigidity). In contrast to the significant reversal of parkinsonian symptoms in the MPTP-lesioned monkeys, balance and gait impairments did not change in monkeys with the isolated PPN lesion after apomorphine dopamine agonist treatment. These findings suggest that selective cholinergic lesioning of the PPN can induce a motor syndrome with prominent postural and gait deficits that is unresponsive to dopaminergic therapy (Table 1).

5.5.2 The cholinergic system and mobility in human PD patients: Recent *in vivo* AChE PET imaging findings also support a role for the PPN in the etiology of PIGD features in PD. In two different patient populations, we showed recently that PD fallers had significantly decreased thalamic cholinergic innervation compared to non-fallers, but did not differ in the degree of nigrostriatal dopaminergic denervation [56, 88]. Thalamic AChE activity derives mainly from terminals of brainstem PPN neurons and loss of thalamic AChE is therefore likely to reflect PPN neuron dysfunction or degeneration. Functional MRI studies have also suggested a role for the PPN, consistently showing activation of this region during motor imagery of gait paradigms [87, 89–91]. Karachi *et al.* have also presented human post-mortem findings showing more severe PPN cholinergic cell losses in PD fallers compared to PD non-fallers [87]. Post-mortem studies have shown greater PPN cholinergic losses in patients with PSP compared to PD [33]. As gait and balance problems are a prominent feature in PSP, the presence of relatively isolated PPN cholinergic degeneration in this disorder provides further evidence for an important role of the PPN in postural control, as it is plausible that the much higher incidence of falls in PSP compared to PD may reflect the more prominent degree of cholinergic PPN pathology in PSP.

We recently reported that BF cholinergic degeneration is associated with slower walking in patients with PD [56]. However, these findings were not corrected for the degree of nigrostriatal denervation and no comparison was made with non-PD control subjects. Recent

preliminary findings confirm that BF cholinergic denervation is associated with slower walking while findings remain significant when corrected for the degree of nigrostriatal denervation, age, cognition, and duration of disease. Further analysis of the preliminary data shows also that walking is not significantly slower than in normal control subjects in the subgroup of PD subjects with mainly nigrostriatal dopaminergic denervation and normal-range cholinergic innervation [92]. These findings suggest that BF cholinergic denervation may be a possible marker of slowing of gait in PD. However, the exact mechanism of the cortical cholinergic system in the control of gait is not known but may be related to decreased cognitive processing abilities during ambulation [93]. These observations may explain also why patients with more severe PIGD features in PD are at an increased risk of developing dementia.

6. Future directions

Multisystem neurodegeneration may play an important role in the etiology of dopamineresistant non-motor and motor symptoms of PD. While there is nigrostriatal dopaminergic denervation in all clinically phase PD patients, our findings of heterogeneity of cholinergic denervation suggests that there are PD patients with relatively normal cholinergic innervation (“single-system pathology”) versus those with additional degeneration of the cholinergic system (“multisystem pathology”). Both from a research and a clinical perspective it may become increasingly more important to correctly identify patients with multisystem degenerations in PD. Dual-tracer dopaminergic and cholinergic PET studies will allow objective classification of PD patients into the presumed single and multisystem degeneration categories in a research setting. Although PET imaging may not be practical in a clinical setting, proxy measures of cholinergic system integrity may provide physiological or clinical markers to identify the subgroup of patients with more prominent cholinergic losses.

6.1 Proxy measures of cholinergic system integrity—Recently, Yarnall, Burn, Rochester, and their colleagues used short latency afferent inhibition of evoked potentials by using transcranial magnetic stimulation of the motor cortex as a proxy measure of cholinergic system integrity. Results of their studies showed that there was no significant difference between cognitively normal PD and control subjects for short latency afferent inhibition. However, PD subjects with mild cognitive impairment had significantly less inhibition compared to both cognitively normal PD and controls [94]. Using this same methodology they also reported that cortical cholinergic dysfunction may be an important contributor to early gait dysfunction in PD [95].

There is a need to identify and validate simple clinical measures that may be markers of cholinergic system loss and could be used to identify suitable patients for future targeted cholinergic drug treatment in PD (so-called “personalized medicine”).

6.2 Adaptive brain plasticity in PD—Multisystem pathologies in PD may herald more severe and disabling clinical symptoms. The relatively isolated impairment of dopaminergic single-system pathology manifests as the typical impairments of PD observed in a newly diagnosed patient, i.e. asymmetric upper limb bradykinesia, mild rigidity, tremor, reduced

arm swing but generally good walking speed and balance. Gait and balance impairments in PD probably result from an intricate interplay of multi-system degenerations and neurotransmitter deficiencies. We hypothesize that gait and postural impairments may be mild in early PD because of adaptive plasticity in the remaining intact non-dopaminergic systems. However, once PD advances to the multisystem stage of disease, the initially compensating non-dopaminergic systems cannot adapt further and more severe clinical morbidity becomes manifest. In this respect, cholinergic system denervation may manifest with gait slowing and increased fall risk. It should be noted, however, that cholinergic system denervation is only one of other possible degenerations contributing to increasing feature severity in multisystem pathologies in PD. Post-mortem studies of end-stage PD and *in vivo* imaging studies show evidence of noradrenergic and serotonergic degeneration, and Alzheimer pathology among other degenerations [96–99]. For example, we recently reported an association between the increased presence of cortical β -amyloid depositions and worsening PIGD features in PD [100]. We believe that operationalization of multisystem degeneration underlying mobility functions in PD is a novel approach and may provide a unique framework to study complex disease mechanisms in advancing PD.

Conclusion

We conclude that cholinergic degeneration is an important contributor to a number of clinical features of PD, including cognition, olfaction, PIGD features, presence of RBD, and mood. Evidence is accumulating for roles for both the cholinergic PPN-thalamic complex and BF cortical projections in the etiology of some PIGD features in PD. Cholinergic system degeneration has a variable presence in PD patients without dementia. More readily identification of PD patients with cholinergic system degeneration may possibly allow future targeted cholinergic treatment approaches, in addition to dopaminergic therapy, to ameliorate a diverse spectrum of non-motor and motor clinical morbidity.

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References

References

Papers of particular interest, published recently, have been highlighted as:

• Of importance

•• Of major importance

1. Langston JW. The Parkinson's complex: parkinsonism is just the tip of the iceberg. *Ann Neurol*. 2006; 59:591–596. [PubMed: 16566021]
2. Braak H, Del Tredici K, Rub U, et al. Staging of brain pathology related to sporadic Parkinson's disease. *Neurobiol Aging*. 2003; 24:197–211. [PubMed: 12498954]
3. Bohnen NI, Kaufer DI, Ivancu LS, et al. Cortical cholinergic function is more severely affected in parkinsonian dementia than in Alzheimer disease: an *in vivo* positron emission tomographic study. *Arch Neurol*. 2003; 60:1745–1748. [PubMed: 14676050]

4. Bohnen NI, Albin RL. The cholinergic system and Parkinson disease. *Behav Brain Res.* 2011; 221:564–573. [PubMed: 20060022]
5. Mesulam MM, Geula C. Nucleus basalis (Ch4) and cortical cholinergic innervation in the human brain: observations based on the distribution of acetylcholinesterase and choline acetyltransferase. *J Comp Neurol.* 1988; 275:216–240. [PubMed: 3220975]
6. Heckers S, Geula C, Mesulam MM. Cholinergic innervation of the human thalamus: dual origin and differential nuclear distribution. *J Comp Neurol.* 1992; 325:68–82. [PubMed: 1282919]
7. de Lacalle S, Hersh LB, Saper CB. Cholinergic innervation of the human cerebellum. *J Comp Neurol.* 1993; 328:364–376. [PubMed: 8440786]
8. Fibiger HC. The organization and some projections of cholinergic neurons of the mammalian forebrain. *Brain Res.* 1982; 257:327–388. [PubMed: 6756546]
9. Lecourtier L, Kelly PH. A conductor hidden in the orchestra? Role of the habenular complex in monoamine transmission and cognition. *Neurosci Biobehav Rev.* 2007; 31:658–672. [PubMed: 17379307]
10. Mesulam MM, Mash D, Hersh L, et al. Cholinergic innervation of the human striatum, globus pallidus, subthalamic nucleus, substantia nigra, and red nucleus. *J Comp Neurol.* 1992; 323:252–268. [PubMed: 1401259]
11. Flores CM, Rogers SW, Pabreza LA, et al. A subtype of nicotinic cholinergic receptor in rat brain is composed of $\alpha 4$ and $\beta 2$ subunits and is up-regulated by chronic nicotine treatment. *Mol Pharmacol.* 1992; 41:31–37. [PubMed: 1732720]
12. Court J, Clementi F. Distribution of nicotinic subtypes in human brain. *Alzheimer Dis Assoc Disord.* 1995; (9 Suppl 2):6–14. [PubMed: 8534425]
13. Cortes R, Palacios JM. Muscarinic cholinergic receptor subtypes in the rat brain. I. Quantitative autoradiographic studies. *Brain Res.* 1986; 362:227–238. [PubMed: 3942874]
14. Cortes R, Probst A, Tobler HJ, Palacios JM. Muscarinic cholinergic receptor subtypes in the human brain. II. Quantitative autoradiographic studies. *Brain Res.* 1986; 362:239–253. [PubMed: 3753655]
15. Mesulam M, Shaw P, Mash D, Weintraub S. Cholinergic nucleus basalis tauopathy emerges early in the aging-MCI-AD continuum. *Ann Neurol.* 2004; 55:815–828. [PubMed: 15174015]
16. Kotagal V, Muller ML, Kaufer DI, et al. Thalamic cholinergic innervation is spared in Alzheimer disease compared to parkinsonian disorders. *Neurosci Lett.* 2012; 514:169–172. [PubMed: 22414859]
17. Davis KL, Mohs RC, Marin D, et al. Cholinergic markers in elderly patients with early signs of Alzheimer disease. *JAMA.* 1999; 281:1401–1406. [PubMed: 10217056]
18. DeKosky ST, Ikonomic MD, Styren SD, et al. Upregulation of choline acetyltransferase activity in hippocampus and frontal cortex of elderly subjects with mild cognitive impairment. *Ann Neurol.* 2002; 51:145–155. [PubMed: 11835370]
19. Mufson EJ, Ma SY, Dills J, et al. Loss of basal forebrain P75(NTR) immunoreactivity in subjects with mild cognitive impairment and Alzheimer's disease. *J Comp Neurol.* 2002; 443:136–153. [PubMed: 11793352]
20. Tiraboschi P, Hansen LA, Alford M, et al. The decline in synapses and cholinergic activity is asynchronous in Alzheimer's disease. *Neurology.* 2000; 55:1278–1283. [PubMed: 11087768]
21. Geula C, Mesulam MM. Systematic regional variations in the loss of cortical cholinergic fibers in Alzheimer's disease. *Cereb Cortex.* 1996; 6:165–177. [PubMed: 8670647]
22. Geula C, Mesulam MM. Cortical cholinergic fibers in aging and Alzheimer's disease: a morphometric study. *Neuroscience.* 1989; 33:469–481. [PubMed: 2636703]
23. Schliebs R, Arendt T. The cholinergic system in aging and neuronal degeneration. *Behav Brain Res.* 2011; 221:555–563. [PubMed: 21145918]
24. Holdorff B. Fritz Heinrich Lewy (1885–1950). *J Neurol.* 2006; 253:677–678. [PubMed: 16767545]
25. Candy JM, Perry RH, Perry EK, et al. Pathological changes in the nucleus of Meynert in Alzheimer's and Parkinson's diseases. *J Neurol Sci.* 1983; 59:277–289. [PubMed: 6854353]
26. Nakano I, Hirano A. Parkinson's disease: neuron loss in the nucleus basalis without concomitant Alzheimer's disease. *Ann Neurol.* 1984; 15:415–418. [PubMed: 6732189]

27. Rogers JD, Brogan D, Mirra SS. The nucleus basalis of Meynert in neurological disease: a quantitative morphological study. *Ann Neurol.* 1985; 17:163–170. [PubMed: 3883886]
28. Tagliavini F, Pilleri G, Bouras C, Constantinidis J. The basal nucleus of Meynert in idiopathic Parkinson's disease. *Acta Neurol Scand.* 1984; 70:20–28. [PubMed: 6475484]
29. Whitehouse PJ, Hedreen JC, White CL 3rd, Price DL. Basal forebrain neurons in the dementia of Parkinson disease. *Ann Neurol.* 1983; 13:243–248. [PubMed: 6847136]
30. Arendt T, Bigl V, Arendt A, Tennstedt A. Loss of neurons in the nucleus basalis of Meynert in Alzheimer's disease, paralysis agitans and Korsakoff's Disease. *Acta Neuropathol.* 1983; 61:101–108. [PubMed: 6637393]
31. Jellinger K. The pedunculopontine nucleus in Parkinson's disease, progressive supranuclear palsy and Alzheimer's disease. *J Neurol Neurosurg Psychiatry.* 1988; 51:540–543. [PubMed: 3379428]
32. Gai WP, Halliday GM, Blumbergs PC, et al. Substance P-containing neurons in the mesopontine tegmentum are severely affected in Parkinson's disease. *Brain.* 1991; 114:2253–2267. [PubMed: 1718530]
33. Hirsch EC, Graybiel AM, Duyckaerts C, Javoy-Agid F. Neuronal loss in the pedunculopontine tegmental nucleus in Parkinson disease and in progressive supranuclear palsy. *Proc Natl Acad Sci U S A.* 1987; 84:5976–5980. [PubMed: 3475716]
34. Zweig RM, Jankel WR, Hedreen JC, et al. The pedunculopontine nucleus in Parkinson's disease. *Ann Neurol.* 1989; 26:41–46. [PubMed: 2549845]
35. McKeith IG, Dickson DW, Lowe J, et al. Diagnosis and management of dementia with Lewy bodies: third report of the DLB Consortium. *Neurology.* 2005; 65:1863–1872. [PubMed: 16237129]
36. Zaccai J, Brayne C, McKeith I, et al. Patterns and stages of alpha-synucleinopathy: Relevance in a population-based cohort. *Neurology.* 2008; 70:1042–1048. [PubMed: 18362284]
37. Perry EK, Irving D, Kerwin JM, et al. Cholinergic transmitter and neurotrophic activities in Lewy body dementia: similarity to Parkinson's and distinction from Alzheimer disease. *Alzheimer Dis Assoc Disord.* 1993; 7:69–79. [PubMed: 8347330]
38. Samuel W, Alford M, Hofstetter CR, Hansen L. Dementia with Lewy bodies versus pure Alzheimer disease: differences in cognition, neuropathology, cholinergic dysfunction, and synapse density. *J Neuropathol Exp Neurol.* 1997; 56:499–508. [PubMed: 9143263]
39. Tiraboschi P, Hansen LA, Alford M, et al. Early and widespread cholinergic losses differentiate dementia with Lewy bodies from Alzheimer disease. *Arch Gen Psychiatry.* 2002; 59:946–951. [PubMed: 12365882]
40. Tiraboschi P, Hansen LA, Alford M, et al. Cholinergic dysfunction in diseases with Lewy bodies. *Neurology.* 2000; 54:407–411. [PubMed: 10668703]
41. Wenning GK, Ebersbach G, Verny M, et al. Progression of falls in postmortem-confirmed parkinsonian disorders. *Mov Disord.* 1999; 14:947–950. [PubMed: 10584668]
42. Shinotoh H, Namba H, Yamaguchi M, et al. Positron emission tomographic measurement of acetylcholinesterase activity reveals differential loss of ascending cholinergic systems in Parkinson's disease and progressive supranuclear palsy. *Ann Neurol.* 1999; 46:62–69. [PubMed: 10401781]
43. Gilman S, Koeppe RA, Nan B, et al. Cerebral cortical and subcortical cholinergic deficits in parkinsonian syndromes. *Neurology.* 2010; 74:1416–1423. [PubMed: 20439843]
44. Mesulam MM, Geula C. Overlap between acetylcholinesterase-rich and choline acetyltransferase-positive (cholinergic) axons in human cerebral cortex. *Brain Res.* 1992; 577:112–120. [PubMed: 1521137]
45. Atack JR, Perry EK, Bonham JR, et al. Molecular forms of acetylcholinesterase and butyrylcholinesterase in the aged human central nervous system. *J Neurochem.* 1986; 47:263–277. [PubMed: 3711902]
46. Hilker R, Thomas AV, Klein JC, et al. Dementia in Parkinson disease: functional imaging of cholinergic and dopaminergic pathways. *Neurology.* 2005; 65:1716–1722. [PubMed: 16344512]
47. Kuhl DE, Minoshima S, Fessler JA, et al. In vivo mapping of cholinergic terminals in normal aging, Alzheimer's disease, and Parkinson's disease. *Ann Neurol.* 1996; 40:399–410. [PubMed: 8797529]

48. Shimada H, Hirano S, Shinotoh H, et al. Mapping of brain acetylcholinesterase alterations in Lewy body disease by PET. *Neurology*. 2009; 73:273–278. [PubMed: 19474411]
49. Klein JC, Eggers C, Kalbe E, et al. Neurotransmitter changes in dementia with Lewy bodies and Parkinson disease dementia in vivo. *Neurology*. 2010; 74:885–892. [PubMed: 20181924]
50. Marcone A, Garibotto V, Moresco RM, et al. [¹¹C]-MP4A PET cholinergic measurements in amnesic mild cognitive impairment, probable Alzheimer's disease, and dementia with Lewy bodies: a Bayesian method and voxel-based analysis. *J Alzheimers Dis*. 2012; 31:387–399. [PubMed: 22596267]
51. Pimlott SL, Piggott M, Owens J, et al. Nicotinic acetylcholine receptor distribution in Alzheimer's disease, dementia with Lewy bodies, Parkinson's disease, and vascular dementia: *In vitro* binding study using 5-[¹²⁵I]-A-85380. *Neuropsychopharmacology*. 2004; 29:108–116. [PubMed: 12955099]
52. Kas A, Bottlaender M, Gallezot JD, et al. Decrease of nicotinic receptors in the nigrostriatal system in Parkinson's disease. *J Cereb Blood Flow Metab*. 2009; 29:1601–1608. [PubMed: 19491921]
53. Fujita M, Ichise M, Zoghbi SS, et al. Widespread decrease of nicotinic acetylcholine receptors in Parkinson's disease. *Ann Neurol*. 2006; 59:174–177. [PubMed: 16374823]
54. Meyer PM, Strecker K, Kendziorra K, et al. Reduced $\alpha 4\beta 2^*$ -nicotinic acetylcholine receptor binding and its relationship to mild cognitive and depressive symptoms in Parkinson disease. *Arch Gen Psychiatry*. 2009; 66:866–877. [PubMed: 19652126]
55. Asahina M, Suhara T, Shinotoh H, et al. Brain muscarinic receptors in progressive supranuclear palsy and Parkinson's disease: a positron emission tomographic study. *J Neurol Neurosurg Psychiatry*. 1998; 65:155–163. [PubMed: 9703164]
56. Bohnen NI, Muller ML, Kotagal V, et al. Heterogeneity of cholinergic denervation in Parkinson's disease without dementia. *J Cereb Blood Flow Metab*. 2012; 32:1609–1617. [PubMed: 22569194] This study shows that cholinergic loss is variable in non-demented PD patients. The heterogeneity of cholinergic losses across non-demented patients suggests that cholinergic treatment in these patients should be targeted.
57. Aarsland D, Kurz MW. The epidemiology of dementia associated with Parkinson disease. *J Neurol Sci*. 2010; 289:18–22. [PubMed: 19733364]
58. Koerts J, Leenders KL, Brouwer WH. Cognitive dysfunction in non-demented Parkinson's disease patients: controlled and automatic behavior. *Cortex*. 2009; 45:922–929. [PubMed: 19327762]
59. Sawamoto N, Piccini P, Hotton G, et al. Cognitive deficits and striato-frontal dopamine release in Parkinson's disease. *Brain*. 2008; 131:1294–1302. [PubMed: 18362097]
60. Cools R. Dopaminergic modulation of cognitive function-implications for L-DOPA treatment in Parkinson's disease. *Neurosci Biobehav Rev*. 2006; 30:1–23. [PubMed: 15935475]
61. Ruberg M, Rieger F, Villageois A, et al. Acetylcholinesterase and butyrylcholinesterase in frontal cortex and cerebrospinal fluid of demented and non-demented patients with Parkinson's disease. *Brain Res*. 1986; 362:83–91. [PubMed: 3942870]
62. Mattila PM, Roytta M, Lonnberg P, et al. Choline acetyltransferase activity and striatal dopamine receptors in Parkinson's disease in relation to cognitive impairment. *Acta Neuropathol*. 2001; 102:160–166. [PubMed: 11563631]
63. Bohnen NI, Kaufer DI, Hendrickson R, et al. Cognitive correlates of cortical cholinergic denervation in Parkinson's disease and parkinsonian dementia. *J Neurol*. 2006; 253:242–247. [PubMed: 16133720]
64. Kehagia AA, Barker RA, Robbins TW. Cognitive impairment in Parkinson's disease: the dual syndrome hypothesis. *Neurodegener Dis*. 2013; 11:79–92. [PubMed: 23038420]
65. Williams-Gray CH, Evans JR, Goris A, et al. The distinct cognitive syndromes of Parkinson's disease: 5 year follow-up of the CamPaIGN cohort. *Brain*. 2009; 132:2958–2969. [PubMed: 19812213]
66. Aubert I, Araujo DM, Cecyre D, et al. Comparative alterations of nicotinic and muscarinic binding sites in Alzheimer's and Parkinson's diseases. *J Neurochem*. 1992; 58:529–541. [PubMed: 1729398]

67. Whitehouse PJ, Martino AM, Wagster MV, et al. Reductions in [3H]nicotinic acetylcholine binding in Alzheimer's disease and Parkinson's disease: an autoradiographic study. *Neurology*. 1988; 38:720–723. [PubMed: 3362368]
68. Petrou M, Kotagal V, Bohnen NI. An update on brain imaging in parkinsonian dementia. *Imaging Med*. 2012; 4:201–213. [PubMed: 22768021]
69. Kulisevsky J, Pagonabarraga J, Pascual-Sedano B, et al. Prevalence and correlates of neuropsychiatric symptoms in Parkinson's disease without dementia. *Mov Disord*. 2008; 23:1889–1896. [PubMed: 18709682]
70. Bohnen NI, Kaufer DI, Hendrickson R, et al. Cortical cholinergic denervation is associated with depressive symptoms in Parkinson's disease and parkinsonian dementia. *J Neurol Neurosurg Psychiatry*. 2007; 78:641–643. [PubMed: 17507447]
71. Troster AI, Stalp LD, Paolo AM, et al. Neuropsychological impairment in Parkinson's disease with and without depression. *Arch Neurol*. 1995; 52:1164–1169. [PubMed: 7492290]
72. Mayeux R, Stern Y, Rosen J, Leventhal J. Depression, intellectual impairment, and Parkinson disease. *Neurology*. 1981; 31:645–650. [PubMed: 7195481]
73. Lieberman A. Are dementia and depression in Parkinson's disease related? *J Neurol Sci*. 2006; 248:138–142. [PubMed: 16814323]
74. Weintraub D, Morales KH, Moberg PJ, et al. Antidepressant studies in Parkinson's disease: a review and meta-analysis. *Mov Disord*. 2005; 20:1161–1169. [PubMed: 15954137]
75. Haehner A, Boesveldt S, Berendse HW, et al. Prevalence of smell loss in Parkinson's disease--a multicenter study. *Parkinsonism Relat Disord*. 2009; 15:490–494. [PubMed: 19138875]
76. Doty RL. Olfactory dysfunction in Parkinson disease. *Nat Rev Neurol*. 2012; 8:329–339. [PubMed: 22584158]
77. Bohnen NI, Muller ML. In vivo neurochemical imaging of olfactory dysfunction in Parkinson's disease. *J Neural Transm*. 2013; 120:571–576. [PubMed: 23263541]
78. Bohnen NI, Muller ML, Kotagal V, et al. Olfactory dysfunction, central cholinergic integrity and cognitive impairment in Parkinson's disease. *Brain*. 2010; 133:1747–1754. [PubMed: 20413575]
79. Bohnen NI, Gedela S, Herath P, et al. Selective hyposmia in Parkinson disease: association with hippocampal dopamine activity. *Neurosci Lett*. 2008; 447:12–16. [PubMed: 18838108]
80. Marion MH, Qurashi M, Marshall G, Foster O. Is REM sleep behaviour disorder (RBD) a risk factor of dementia in idiopathic Parkinson's disease? *J Neurol*. 2008; 255:192–196. [PubMed: 18217187]
81. Vendette M, Gagnon JF, Decary A, et al. REM sleep behavior disorder predicts cognitive impairment in Parkinson disease without dementia. *Neurology*. 2007; 69:1843–1849. [PubMed: 17984452]
82. Postuma RB, Gagnon JF, Montplaisir J. Cognition in REM sleep behavior disorder - A window into preclinical dementia? *Sleep Med*. 2008; 9:341–342. [PubMed: 17869575]
83. Kotagal V, Albin RL, Muller ML, et al. Symptoms of rapid eye movement sleep behavior disorder are associated with cholinergic denervation in Parkinson disease. *Ann Neurol*. 2012; 71:560–568. [PubMed: 22522445] To the best of our knowledge this is the first in vivo PET study to show that REM sleep behavior disorder (RBD) is associated with cholinergic denervation in PD. RBD is one of the first prodromal symptoms of Lewy body disorders. This result may indicate early cholinergic system degeneration in some patients.
84. Postuma RB, Gagnon JF, Montplaisir JY. REM sleep behavior disorder: from dreams to neurodegeneration. *Neurobiol Dis*. 2012; 46:553–558. [PubMed: 22019718]
85. Muslimovic D, Post B, Speelman JD, et al. Determinants of disability and quality of life in mild to moderate Parkinson disease. *Neurology*. 2008; 70:2241–2247. [PubMed: 18519873]
86. Sethi K. Levodopa unresponsive symptoms in Parkinson disease. *Mov Disord*. 2008; 23(Suppl 3):S521–533. [PubMed: 18781679]
87. Karachi C, Grabli D, Bernard FA, et al. Cholinergic mesencephalic neurons are involved in gait and postural disorders in Parkinson disease. *J Clin Invest*. 2010; 120:2745–2754. [PubMed: 20628197] An elegant study that convincingly shows the role of the PPN in mobility impairment in parkinsonian monkeys and human patients with PD. Evidence for this is described in threefold in complementary human fMRI, human post-mortem, and primate lesioning studies. The finding

that posture and gait deficits can occur with isolated PPN impairment in the absence of nigrostriatal dopaminergic lesions has major implications for current understanding of mobility impairments in PD.

88. Bohnen NI, Muller ML, Koeppe RA, et al. History of falls in Parkinson disease is associated with reduced cholinergic activity. *Neurology*. 2009; 73:1670–1676. [PubMed: 19917989]
89. Bakker M, de Lange FP, Stevens JA, et al. Motor imagery of gait: a quantitative approach. *Exp Brain Res*. 2007; 179:497–504. [PubMed: 17211663]
90. Cremers J, D'Ostilio K, Stamatakis J, et al. Brain activation pattern related to gait disturbances in Parkinson's disease. *Mov Disord*. 2012; 27:1498–1505. [PubMed: 23008169]
91. Snijders AH, Leunissen I, Bakker M, et al. Gait-related cerebral alterations in patients with Parkinson's disease with freezing of gait. *Brain*. 2011; 134:59–72. [PubMed: 21126990]
92. Bohnen N, Kotagal V, Albin R, et al. Gait speed is preserved in oligosystem compared to multisystem neurodegeneration in Parkinson disease. *Neurology*. 2013; 80:P04165.
93. Woollacott M, Shumway-Cook A. Attention and the control of posture and gait: a review of an emerging area of research. *Gait Posture*. 2002; 16:1–14. [PubMed: 12127181]
94. Yarnall AJ, Rochester L, Baker MR, et al. Short latency afferent inhibition: A biomarker for mild cognitive impairment in Parkinson's disease? *Mov Disord*. 2013
95. Rochester L, Yarnall AJ, Baker MR, et al. Cholinergic dysfunction contributes to gait disturbance in early Parkinson's disease. *Brain*. 2012; 135:2779–2788. [PubMed: 22961550]
96. Albin RL, Koeppe RA, Bohnen NI, et al. Sparing caudal brainstem SERT binding in early Parkinson's disease. *J Cereb Blood Flow Metab*. 2008; 28:441–444. [PubMed: 18073772]
97. Maetzler W, Liepelt I, Reimold M, et al. Cortical PIB binding in Lewy body disease is associated with Alzheimer-like characteristics. *Neurobiol Dis*. 2009; 34:107–112. [PubMed: 19162186]
98. Maetzler W, Reimold M, Liepelt I, et al. [¹¹C]PIB binding in Parkinson's disease dementia. *Neuroimage*. 2008; 39:1027–1033. [PubMed: 18035558]
99. Mann DM, Yates PO, Hawkes J. The pathology of the human locus ceruleus. *Clin Neuropathol*. 1983; 2:1–7. [PubMed: 6220852]
100. Muller ML, Frey KA, Petrou M, et al. β -amyloid and postural instability and gait difficulty in Parkinson's disease at risk for dementia. *Mov Disord*. 2013; 28:296–301. [PubMed: 23239424]

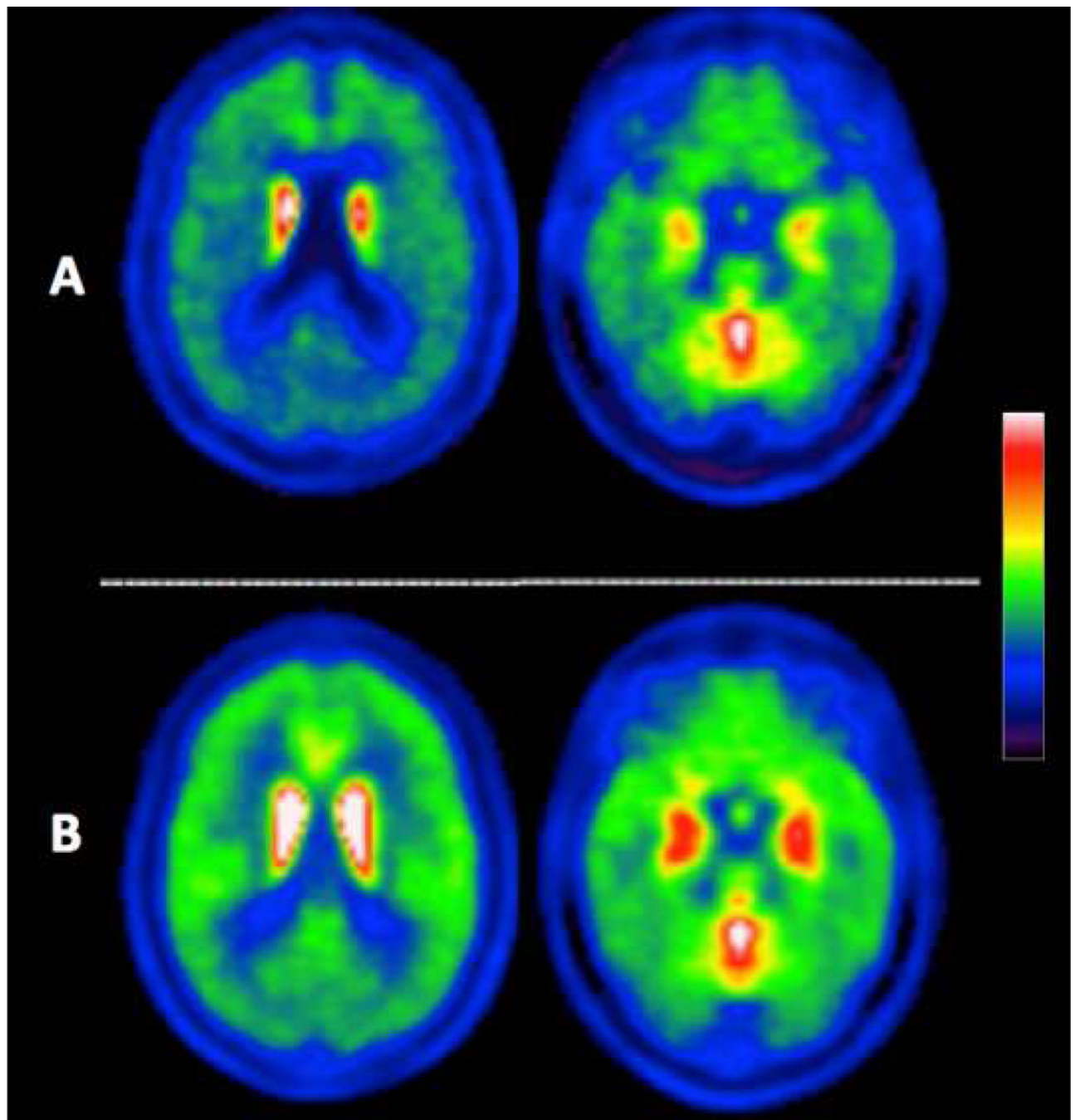


Figure 1. Transaxial images of a vesicular acetylcholine transporter ($[^{18}\text{F}]\text{FEOBV}$) PET study shows widespread cholinergic denervation in a patient with parkinsonian dementia (A: top row) compared to a healthy control subject (B: bottom row).

Table 1

Findings of selective dopamine (DA) and PPN cholinergic (ACh) lesions in rhesus monkeys. Data extracted from Karachi *et al.* [87].

	Nigral DA cells	PPN ACh cells	Postural deficits
Isolated MPTP lesion young monkey	↓↓ DA (~90%)	normal	No
Isolated MPTP lesion old monkey	↓↓ DA (~90%)	↓ ACh (~30%)	Yes
Isolated PPN lesion	Normal	↓ ACh (~30%)	Yes