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Biomarkers of Parkinson's disease: current status and future

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Abstract

This review summarizes major advances in biomarker discovery for diagnosis, differential diagnosis, and progression of Parkinson's disease (PD), with emphasis on neuroimaging and biochemical markers. Potential strategies to develop biomarkers capable of predicting PD in the prodromal stage before the appearance of motor symptoms or correlating with nonmotor symptoms, an active area of research, are also discussed.

Keywords

Parkinson disease; biomarkers; neuroimaging; proteomics; metabolomics; prodromal diagnosis

Parkinson's disease (PD) is a prevalent neurodegenerative disorder, with its motor symptoms largely attributable to the degeneration of dopaminergic (DAergic) neurons. It has become clear, however, that PD is also associated with nonmotor symptoms in conjunction with neuronal loss in many other brain regions that occur before or after the loss of DAergic neurons [1]. The misdiagnosis rate of PD can range from 10–50% by movement disorder specialists [2] due in part to the fact that there are no sensitive and specific biomarkers validated for clinicians to differentiate PD from other movement disorders with overlapping clinical symptoms. Apart from diagnostic utility, biomarkers for PD are also needed for monitoring disease progression and efficacy of interventions, which are currently assessed by severity of motor symptoms.

An ideal PD biomarker should meet the following qualifications: high sensitivity and specificity validated by neuropathological examination, satisfactory test–retest reproducibility, easy accessibility, inexpensive, and offer the ability to monitor disease progression without being biased by age, compensatory mechanisms, or treatments. While no such biomarker to date fulfills all of these criteria for PD, this review will discuss two areas of research, neuroimaging and biochemical markers, which have demonstrated obvious potential in diagnosing the disease and monitoring its progression. Future biomarker

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investigations relating to prodromal biomarkers and markers related to nonmotor symptoms are also discussed.

Neuroimaging biomarkers

To date, the most mature PD biomarkers for nigrostriatal neurodegeneration are those employing neuroimaging methodologies. Currently, controversy surrounds whether these techniques can be effective in differentiating clinically overlapping parkinsonisms and/or objectively assessing PD progression. The major methods used as well as their current clinical and research utility are discussed below and summarized in Table 1.

Dopaminergic imaging

Aromatic amino acid decarboxylase—6-[^{18}F]-fluoro-1-3,4-dihydroxyphenylalanine (^{18}F -dopa) positron emission tomography (PET) was the first neuroimaging approach validated for measuring and assessing presynaptic DAergic neuronal integrity. To reach the striatum, ^{18}F -dopa is transported across the blood–brain barrier, taken up by axonal terminals of DA neurons, decarboxylated by aromatic amino acid decarboxylase (AADC), converted into ^{18}F -DA, and stored in vesicles. The key determinants for ^{18}F -dopa uptake are the density of the axonal terminal plexus and AADC activity, which reflect the number of remaining nigral DAergic cells. Patients with early PD show 50% decreased ^{18}F -dopa uptake in the putamen [3], with the greatest reduction occurring in a gradient pattern in the posterior dorsal putamen contralateral to the side of symptom onset [4,5]. Although such imaging appears to be effective in confirming symptomatic PD, its utility in assessing PD progression [6,7] and detecting prodromal PD are questionable because ^{18}F -dopa PET may underestimate the degree of degeneration due to compensatory upregulation of AADC in remaining terminals [8].

Dopamine transporter—The dopamine transporter (DAT), a protein expressed on the membrane of presynaptic DA terminals and involved in the reuptake of DA, can be assessed with PET using several tracers, including ^{11}C -CFT, ^{18}F -FP-CIT, ^{11}C -RTI-32, and ^{11}C -methylphenidate. Single photon emission computed tomography (SPECT), a technology used routinely in clinical practice, can also be used to image DAT through ^{123}I -b-CIT, ^{123}I -FP-CIT, ^{123}I -IPT, ^{123}I -altropine, or $^{99\text{m}}\text{Tc}$ -TRODAT-1 [9]. DAT imaging is used as an *in vivo* marker, reflecting integrity and number of DA neurons. A few studies have reported significantly reduced striatal DAT in more than 95% of parkinsonism cases [10], including those at early stages [11].

With respect to monitoring PD progression and efficacy of putative agents, the value of DAT imaging remains to be established. Although some investigations suggest that striatal uptake of ^{18}F -FP-CIT correlates with Hoehn and Yahr (H&Y) score [12], other clinical trials show that DAT imaging density declined faster in patients with PD treated with levodopa, despite improved clinical motor scores [13]. Symptomatic therapy may therefore influence imaging results in their relation to clinical diagnosis.

Vesicular monoamine transporter 2—Vesicular monoamine transporter 2 (VMAT2), a membrane protein that transports monoamines from the cytosol into secretory vesicles in monoaminergic neurons, is exclusively expressed in the brain and has an essential role in DA reuptake. VMAT2 is imaged using ^{11}C - or ^{18}F -dihydrotetrabenazine (DTBZ) PET and is the newest approach for the assessment of nigrostriatal projections [14]. In patients with PD, striatal ^{11}C -DTBZ is significantly reduced with the putamen exhibiting the greatest decrease [14]. VMAT2 imaging appears to be less sensitive to compensation and pharmacologic regulation [15], giving it the potential to provide the most reliable measurement of the density of DAergic terminals in PD. Nevertheless, the application

of ^{11}C -DTBZ is limited because its short half-life requires a cyclotron on-site [16]. A novel ^{18}F -labeled tetrabenazine derivative, ^{18}F -fluoropropylidihydrotetrabenazine, is currently under development and is showing promise as a longer-lived and lower-cost alternative [16].

Caveats and potential solutions—Among the three techniques discussed above, DAT imaging is the most commonly used to date. However, several limitations associated with its use should be emphasized. DAT imaging likely overestimates the reduction in terminal density in early PD due to compensatory downregulation in remaining neurons. Also, current DAT radioligands bind with other monoamine transporters, particularly serotonin (SERT), which is especially problematic as SERT is the dominant monoamine transporter in the midbrain [17]. Lastly, DAT imaging cannot reliably differentiate between PD and other forms of parkinsonism [18], making the production of additional methods in conjunction with DAT imaging necessary to define a PD biomarker. Recently, fluorine-18 labeled 2- β -carbomethoxy-3 β -(4-chlorophenyl)-8-(2-fluoroethyl)-nortropane (^{18}F -FECNT) was developed as a novel PET tracer with high affinity for DAT and a much lower affinity for the norepinephrine transporter and SERT. Compared with current PET DAT tracers, ^{18}F -FECNT has higher test–retest reproducibility and may be able to track striatal and nigral DA denervation [19], making it potentially useful for longitudinal evaluation of PD progression.

As previously mentioned, another shortcoming of DAergic neuroimaging is that the techniques assess nigrostriatal DA function, rather than true pathology, rendering the obtained values subject to compensatory mechanisms, in which there is upregulation of ^{18}F -dopa uptake and downregulation of DAT binding [15]. To partially circumvent this problem, postsynaptic DA receptors can be examined by the PET ligand ^{11}C -raclopride for $\text{D}_{2/3}$ receptor [20] or SPECT tracer ^{123}I -iodobenzamide for D_2 receptors. Increased levels of D_2 receptor availability can be observed in early stages of *de novo* PD, which is useful in differentiating PD from atypical parkinsonisms [21]. Nonetheless, whether these new targets are truly unaffected by compensatory modulations or medications remains to be investigated.

Non-dopaminergic imaging

An active area of research in neuroimaging markers is assessment of brain functions and structures beyond nigrostriatal DAergic degeneration. These include Parkinson disease-related spatial covariance pattern (PDRP), cholinergic function imaging, and magnetic resonance imaging (MRI). Such strategies incorporate the fact that PD affects non-nigrostriatal DAergic regions of the brain. Transcranial sonography (TCS), also a non-dopamine imaging method, will be discussed in the context of prodromal diagnosis, where utility of imaging methods will be integrated with biochemical markers.

Metabolism network imaging in brain—Investigation with ^{18}F -FDG PET revealed that PD is associated with a specific metabolic network characterized by increased pallidothalamic and pontine metabolism associated with metabolic reductions in the lateral premotor and posterior parietal cortical regions [22,23], termed PDRP. In one study, PDRP network analysis classified idiopathic PD with 84% sensitivity and 97% specificity, helping differentiate PD from atypical parkinsonisms such as multiple system atrophy (MSA) and progressive supranuclear palsy (PSP) [24]. Furthermore, PDRP activity precedes the appearance of motor symptoms by approximately 2 years [25], indicating its potential usefulness in diagnosing PD at its prodromal stage. These facts, coupled with its high level of reproducibility [26] and correlation with clinical severity [27], make PDRP a promising biomarker to track PD progression, monitor therapeutic intervention [28], and could potentially fill voids left by nigrostriatal imaging methods mentioned above.

Cholinergic dysfunction imaging—In addition to PDRP, imaging brain regions that are not part of the nigrostriatal tract shows some promise in diagnosing and distinguishing PD from other neurological diseases. New imaging techniques for assaying cortical acetylcholinesterase activity with ^{11}C -MP4A PET reveal a deficit of cholinergic function throughout the cortex in parallel with the loss of striatal DAergic function in PD [29]. This observation is significant, especially with the realization of nonmotor components in PD (e.g. cognitive impairment) which is intimately associated with the cholinergic system.

MRI—MRI is a form of neuroimaging that is particularly useful in ruling out secondary causes of parkinsonism due to the fact that it is capable of detecting abnormalities in the structure of various brain regions. Recent advances in high-field MRI technology have been increasingly employed in diagnosing PD and are more sensitive towards demonstrating iron deposits in the midbrain of early PD patients [30]. Additionally, diffusion tensor imaging (DTI) for evaluating regional fractional anisotropy has shown changes in the olfactory tract [31], which could be related to anosmia or hyposmia (a symptom that appears in the prodromal stage – see discussion below), and reduction in the nigra of patients with PD [32] which inversely correlates with H&Y score [33].

Biochemical biomarkers

Biochemical markers, especially those reflecting PD pathogenesis, are sorely needed in samples that are readily accessible clinically [e.g. cerebrospinal fluid (CSF), blood, and saliva]. To date, the most extensively tested candidate biochemical markers are those present in CSF and typically stem from genetic investigations which link the candidate to PD development. A few major biochemical markers and their current clinical and research utility are listed in Table 2. However, none of these markers completely fulfill the criteria defined earlier as an ideal biomarker for PD. Most new candidate markers are discovered by ‘-omics’ technology, and are generally in a preliminary stage with the results obtained in small cohorts using cross-sectional study designs. Validation is therefore needed using large cohorts, particularly those with samples collected longitudinally. Finally, similar to the neuroimaging field, major challenges to the biochemical marker field also include defining prodromal biomarkers and those related to nonmotor dysfunctions.

α -Synuclein, DJ-1, and tau

α -Synuclein has been intensely researched as a PD biomarker due to the fact that it is a key protein in Lewy bodies, the pathological hallmark of PD, and mutations or multiplication of its gene are known to cause familial PD [34,35]. α -Synuclein has been reported to be decreased in CSF from patients with PD compared with controls in most well-controlled investigations [36–39], although it does not appear to be able to distinguish between various synucleinopathies independently [39]. One study has reported an inverse relationship between α -synuclein levels and H&Y score [36], indicating measuring α -synuclein levels may not only be useful as a biomarker of PD but also of disease progression. However, the latter observation has not been replicated in most recent studies [38]. Additionally, oligomeric forms of α -synuclein have been found to be elevated in the CSF of patients with PD as compared with controls. When only oligomers were measured, the sensitivity and specificity were calculated to be 75% and 87.5%, respectively, which increases to 89.3% and 90.6% when the ratio of oligomers and/or total α -synuclein is calculated [40]. Similar elevations have also been observed in plasma [41]. A more recent study reports that the CSF level of phosphorylated α -synuclein (PS-129) appears to be more effective than native α -synuclein in differentiating PD from MSA and PSP and correlates with disease severity as assessed using the Unified Parkinson’s Disease Rating Scale (UPDRS) [42], making it a potential candidate to complement the neuroimaging methods discussed above.

DJ-1 is an extensively studied antioxidative protein which, like α -synuclein, is implicated in PD pathogenesis as mutation of its *PARK7* gene results in familial autosomal recessive forms of the disease. Some reports indicate that DJ-1 is increased in CSF from patients with PD compared to controls [43], a finding which was replicated in plasma and extended to show that DJ-1 levels correlate with disease stage as approximated by H&Y scores [44]. Other studies, however, contradict this conclusion by showing DJ-1 to be decreased in CSF from patients with PD compared with controls, and show an apparent age-dependent increase in DJ-1 levels [38]. The discrepancy might be related to methodological variations or contamination of CSF by blood, which contains a comparatively high level of DJ-1 [45].

In addition to α -synuclein [46,47], genome-wide association studies have found associations between the gene encoding tau (*MAPT*) and PD. However, this observation has largely arisen in cohorts of European descent [46] and was not found in a Japanese cohort, indicating there may be geographical differences in genes associated with PD [47]. Studies with larger cohorts have reported that CSF levels of tau and phospho-181 tau (p-tau) are decreased in symptomatic PD compared with controls [48,49].

In addition to the cardinal motor indicators of PD, patients also experience significant nonmotor symptoms. As such, biomarker investigations have been launched in an attempt to characterize these effects. One such symptom is cognitive impairment, for which a decrease in CSF amyloid- β ($A\beta$) levels has been well established in patients with Alzheimer disease (AD). In CSF samples obtained from patients with PD, the $A\beta$ isoforms $A\beta_{40}$ and $A\beta_{42}$ have been found to be decreased as compared with controls. However, in contrast to AD, there is no consistent increase in tau in conjunction with decreased $A\beta$ in patients with PD with cognitive impairment or dementia [48,50,51].

It should be pointed out that due to the heterogeneity of patients with PD, a combination of several markers may be necessary to achieve high sensitivity and specificity. This is evidenced by the fact that the ratio of α -synuclein to the percentage of p-tau (p-tau/tau) is able to distinguish PD from MSA [49]. Furthermore, the ratio of fractalkine, an inflammatory mediator of microglia, to $A\beta_{42}$ positively correlates with PD severity and progression in cross-sectional and longitudinal CSF samples, respectively [49]. Using combinations of protein markers and/or imaging techniques could therefore produce a biomarker that not only diagnoses PD, but also monitors disease progression. Alternatively, markers may also be stratified based on the predominant features of PD, such as tremor or rigidity, or at-risk traits and/or clinical features (discussed below).

Use of 'omics' in biomarker development

A variety of profiling techniques have been employed to find novel markers that could facilitate diagnosis and monitoring of PD progression. Indeed, the use of '-omics' technologies have enabled for high-throughput studies of metabolites, genes, and proteins in the context of comparing patients with PD with controls. This section will discuss the major findings of metabolomic, genomic, gene expression profiling, and proteomic studies aimed at developing PD biomarkers.

Metabolomics—Metabolomics is used to study the profile of small molecules and has been used in a limited capacity to observe differences in metabolites that may aid in PD diagnosis. Previous studies report decreased uric acid and increased glutathione in plasma from patients with PD compared with controls [52]. However, the reduced form of glutathione has been reported to be decreased in CSF of patients with Lewy body disease, with patients with PD showing a non-statistically significant decrease of this metabolite [53]. By contrast, decreased plasma levels of uric acid have been found in both idiopathic PD and PD caused by mutations in leucine rich repeat kinase 2 (*LRRK2*), indicating

decreased uric acid levels may be a universal feature of patients with PD [54]. Given both higher serum and CSF urate concentrations at baseline are associated with slower rates of clinical deterioration [55] and serum urate is reported to decrease with disease progression as measured by H&Y score [56], urate may be important in predicting and monitoring PD. Notably, the LRRK2 study discussed above also calculated the scores of projection to latent structures-discriminant analysis (PLS-DA) for related and unrelated controls in addition to for patients with idiopathic and *LRRK2* PD and found that each form was distinguishable from the control in addition to each other, although common signatures are also noted. Another study using PLS-DA analysis has identified pyruvate as a key metabolite that can distinguish patients with PD from controls [57]. While metabolomic studies may provide a biomarker that can accurately diagnose PD, the limited number of published studies highlights the need for validation as well as comparisons of PD to other neurodegenerative diseases.

Genomic and expression profiles—A majority of the genomic work done relating to PD biomarker research has found the genes *SNCA* and *MAPT*, encoding for α -synuclein and tau, respectively, are associated with PD as described above [46,47]. A variety of studies have also examined differences in mRNA expression between PD and control subjects. Unfortunately, most of these have been performed in brain tissue, making them impractical towards the development of a clinically useful biomarker. There have been a select number of studies that measured transcript differences in blood. One such study looked at the blood mRNA signature and reported a molecular marker of eight genes that are indicative of a greater risk of PD, in particular decreased expression of *ST13* [58]. Results for the expression of *ST13*, however, were not replicated in a separate study [59] and no differences were observed in a similar study of *GSK3B* in blood [60]. By contrast, several other genes discovered initially, including *HIP2* and *HSPA9* [58], appear to be reproducible in other investigations [61,62].

Proteomic profiling—Many studies have profiled various proteomes of PD and other neurodegenerative patients with the goal of detecting differences in proteins between these groups. A commonly used biofluid is CSF, because it is in close contact with the location of primary PD pathology and can be reasonably obtained. One such study used proteomic profiling of CSF from patients with PD, AD, and dementia with Lewy bodies and found unique changes for each group compared with healthy controls among the 1500 (approximately) proteins identified [63]. Additionally, each group was distinguishable from one another with 95% sensitivity achieved [63]. Later studies validated that levels of brain-derived neurotrophic factor, interleukin 8, vitamin D binding protein, β_2 -microglobulin, haptoglobin, apolipoprotein AII, apoE, tau, and A β 42 could accurately classify 90 of 95 healthy controls, 36 of 48 AD patients, and 38 of 40 PD patients when compared with expert diagnosis [64]. This further supports the concept that using multiple proteins may be a key factor in developing a biomarker for PD. Other profiling studies have been performed and have found different candidate markers [65,66], illustrating that reproducibility is low when employing general profiling methods across diverse cohorts.

Future directions of biomarkers in PD

A biomarker that is able to diagnose PD before the onset of motor symptoms would provide a better chance to develop interventions capable of arresting or slowing disease progression. To this end, two types of high risk populations, subjects with genetic mutations leading to familial PD and those with clinical symptoms associated with a high conversion rate to PD, are candidates wherein prodromal biomarkers can be potentially identified. Additionally, discovery of biochemical markers is shifting from CSF to other peripheral biofluids that are

more easily accessible. Imaging, when used in combination with biochemical biomarkers, may allow for the discovery of a specific profile that can predict PD onset.

Prodromal diagnosis

Tools used for prodromal diagnosis—The use of neuroimaging has shown the greatest promise in developing prodromal markers for PD. Nigrostriatal DA imaging, the best established marker of motor dysfunction thus far, should be considered the ‘gold-standard’ in defining subjects at a higher risk for PD before the onset of motor symptoms (discussed below). Additionally, TCS, which is readily available in most clinics, may be useful towards prodromal diagnosis of PD. To this end, TCS has been used to observe the lateral midbrain and studies report increased echogenicity (‘hyperechogenicity’) in the lateral midbrain in approximately 90% of cases [67]. Compared with the clinical standard, diagnosing PD at baseline by TCS was assessed with a sensitivity of 91% and specificity of 82% [68]. TCS may be useful in developing a prodromal biomarker as one study reports 14 of 39 patients with rapid eye movement behavior disorder (RBD), a group of people at risk of PD (discussed below), showed hyperechogenicity in the nigra [69]. Additionally, for individuals 50 years or older without evidence of PD, the relative risk for incident PD in those with enlarged substantia nigra hyperechogenicity was 17 times higher compared with normoechogenic controls after 37 months of prospective follow up [70]. It should be noted, however, the size of the TCS signal did not show changes with disease progression when assessed using H&Y or UPDRS scores [71], suggesting it is not an appropriate biomarker for follow-up evaluation of disease severity.

Population at high risk—To develop a prodromal biomarker for PD, individuals who are at risk of developing PD can be studied before disease onset. Two such populations include individuals who have a genetic risk for PD and people who show symptoms that appear before disease onset. Among individuals with a genetic risk for PD, those with mutations in *LRRK2* or glucocerebrosidase (*GBA*) are worth discussing, because their prevalence is relatively high. Studies on asymptomatic *LRRK2* carriers are likely to be especially informative due to the high penetrance rate of *LRRK2* mutations [72]. DAT imaging of two asymptomatic *LRRK2* mutation carriers showed reduced ^{11}C -MP binding but normal ^{18}F -dopa uptake in putamen [73], which is consistent with decreased binding of DAT and increased activity of dopa decarboxylase in prodromal stages of PD. Additionally, *LRRK2* patients were similar to patients with sporadic PD [73]. A separate study reported a greater than expected decline in PET markers (most commonly ^{11}C -DTBZ and ^{11}C -methylphenidate but also ^{18}F -dopa uptake) for some non-symptomatic *LRRK2* carriers [74], indicating PET imaging before clinical PD onset may be useful in diagnosing PD. Others have shown that *GBA* mutation carriers have decreased cerebral rates of glucose metabolism in the supplemental motor area [75]; further validating the concept that imaging before the appearance of motor symptoms may be an important aspect of a prodromal PD biomarker.

Imaging subjects displaying nonmotor symptoms that commonly occur before the onset of motor symptoms in PD may also be useful in defining a prodromal biomarker. Impaired olfactory function is a common finding in PD patients, occurring early in the course of the disease [76] and may be able to distinguish PD from other movement disorders [77]. Similarly, studies have shown that RBD may precede the onset of parkinsonism [78] in addition to other prodromal symptoms of PD [79], which include depression, constipation, and cardiac dysfunction (the foundation of cardiac scan, see Table 1). DAT imaging has been especially useful towards developing such a biomarker. In a cohort of 361 asymptomatic relatives of patients with PD, idiopathic olfactory dysfunction was assessed and nigrostriatal DA neuron function was evaluated through SPECT DAT imaging. A total of 12.5% of the hyposmic first-degree relatives of patients with PD eventually developed PD

and all had an abnormal baseline SPECT scan [80]. RBD patients similarly have shown reduced striatal DAT binding using ^{123}I -IPT SPECT [69,81], and to a lesser extent FP-CIT SPECT [82], indicating that neuroimaging, particularly DAT imaging, is capable of detecting prodromal DA dysfunction in subjects who are at risk of PD but have not yet developed motor symptoms, as mentioned above. This is indeed confirmed in our recent investigation (Figure 1). Longitudinally monitoring patients that have these conditions and observing the differences between those that develop PD and controls may provide valuable insight into prodromal markers of PD in addition to markers capable of monitoring disease progression.

Patients with *LRRK2* or *GBA* mutations or with prodromal PD symptoms can also be used to develop biochemical biomarkers. Most of the work in this area has focused on *LRRK2* carriers. Plotting PLS-DA scores calculated from asymptomatic *LRRK2* carriers and family members without a mutant copy of *LRRK2* showed that metabolomic profiles of these groups were distinguishable from each other [54]. Additionally, decreased levels of A β and tau species in CSF correlate with decreased DA neuron function in *LRRK2* mutation carriers as detected by imaging through ^{18}F -dopa, ^{11}C -DTBZ and ^{11}C -methylphenidate [83], adding strength to their usefulness as PD biomarkers. Because asymptomatic carriers represent a prodromal state of PD, such findings indicate metabolomics and protein levels may be useful in the development of prodromal biomarkers for PD. Unfortunately, attempts at using α -synuclein or DJ-1 in *LRRK2* patients for the same purpose has not proved successful [84], warranting further investigation into candidate markers in at-risk populations. Longitudinal studies on such populations will likely allow for the development of a prodromal biomarker for PD.

Transition from CSF to peripheral fluids

Although CSF biomarkers have shown great potential for PD diagnosis, CSF is obtained through lumbar puncture, which is relatively invasive compared with obtaining other biofluids such as blood or saliva. Although some studies in plasma have yielded promising results (discussed above) [40,41], further validation is still necessary. One recent study identified α -synuclein and DJ-1 in saliva [85], opening the door for using saliva as a biofluid in which to develop a biomarker for PD. Future studies should aim to integrate neuroimaging techniques in addition to markers in biofluids to develop a marker that is not only specific for PD but also allows for preclinical diagnosis and monitoring of disease progression.

Concluding remarks

The current field of PD biomarkers focuses on using neuroimaging in addition to biochemical markers that can be measured in biofluids. Although a perfect biomarker has not been developed, significant progress has been made towards discovering molecular and imaging patterns that can accurately distinguish PD and monitor its progression. Among imaging methods that are currently available, DAT appears to be most widely used towards diagnosing PD, including defining PD at prodromal stages. However, newer imaging methods are needed to improve differential diagnosis of overlapping parkinsonian disorders and assessing PD progression objectively. Biochemical markers are important not only towards filling the gaps associated with imaging fields, but also in revealing novel molecular targets involved in PD development and progression. Additionally, biochemical markers are typically determined in human fluids, making them advantageous in the aspect that they are easily accessible for application in a routine clinical setting. Future efforts should look to integrate these two fields and extend studies to the prodromal stages of PD in addition to nonmotor symptoms. Correlating imaging data with biochemical markers will likely improve the diagnostic accuracy of PD, including at prodromal stages, in addition to

monitoring its progression and the efficacy of treatment, especially at early phases when neuroprotective therapies are most effective.

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Highlights

- Parkinson's disease has appreciable rate of misdiagnosis.
- Imaging and biochemical markers aim to assist with diagnosis and progression.
- Imaging is more advanced, while biochemical markers may reflect pathogenesis.
- Major areas of research are geared towards developing pre- or non-motor biomarkers.
- Combining imaging and biochemical markers is likely to move the field effectively.

Figure 1. Imaging of dopamine transporter in different types of RBD patients
(a) Idiopathic RBD with normal DAT imaging and no signs for PD (male, 66 yrs); (b) Idiopathic RBD with reduction of DAT in imaging but no signs for PD; however, PD was confirmed clinically after follow up with left limbs affected at onset (male, 70 yrs); (C) Idiopathic RBD with reduction of DAT in imaging and PD (male, 71 yrs).

Table 1

Neuroimaging biomarker candidates and their potential utilities in PD.

Imaging target	Prodromal diagnosis	Confirming PD at its onset	Differentiation from atypical Parkinsonism	Monitoring progression	Sensitivity and specificity for PD diagnosis [Refs]
AADC (¹⁸ F-dopa)	Probably useful	Useful	Unlikely useful	Possibly useful	
DAT	Useful	Useful	Unlikely useful	Possibly useful	98% and 83% [86], 92% and 100% [18]
VMAT2	Probably useful	Useful	Investigational	Investigational	
FDG network	Probably useful	Probably useful	Probably useful	Possibly useful	95% and 94% [87]
Dopamine receptors	Non-useful	Non-useful alone	Non-useful alone	Non-useful	
Cholinergic function	Investigational	Investigational	Probably useful (for DLB)	Investigational	
Cardiac autonomic dysfunction	Investigational	Non-useful	Useful (for MSA)	Investigational	
TCS	Probably useful	Useful	Investigational	Unlikely useful	91% and 82% [68]
MRI (DWI, iron deposit)	Investigational	Probably useful	Investigational	Investigational	

Table 2

Biochemical Biomarker candidates and their potential utilities in PD.

Biochemical marker(s)	Prodromal diagnosis [Refs]	Confirming PD at its onset [Refs]	Differentiation from atypical Parkinsonism [Refs]	Monitoring progression [Refs]	Sensitivity and specificity for PD diagnosis [Refs]
α -Synuclein	Unlikely in LRRK2 cases [83]	Possibly useful [36–39]	Possibly useful-potentially more useful if PS-129 is measured or it is used with the ratio of p-tau/tau [42],	Possibly useful [42]	Measuring oligomers 75% and 87.5%; improves to 89.3% and 90.6% when measuring the ratio of oligomers:total [40]
DJ-1	Possibly useful in G2019S LRRK2 cases [83]	Possibly useful [43]	Unknown	Possibly useful [43]	
A β	Possibly useful [83]	Possibly useful [48–50]	Unknown	Possibly useful if used in conjunction with other protein(s), e.g. tau, fractalkine [48–50].	
Tau	Possibly useful [83]	Possibly useful [48,49]	Possibly useful if used in a p-tau/tau ratio with α -synuclein [42]	Possibly useful if used in conjunction with A β [48,50,51]	
Uric Acid	Unknown	Possibly useful [52]	Unknown	Possibly useful [55]	
Glutathione	Unknown	Possibly useful [52, 53]	Unknown	Unknown	