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REVIEW ARTICLE

Emerging urinary alpha-synuclein and miRNA biomarkers in Parkinson's disease

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Abstract

Parkinson's disease (PD) is one of the most common neurodegenerative diseases after Alzheimer's disease (AD), afflicting adults above the age of sixty irrespective of gender, race, ethnicity, and social status. PD is characterized by motor dysfunctions, displaying resting tremor, rigidity, bradykinesia, and postural imbalance. Non-motor symptoms, including rapid eye movement (REM) behavior disorder, constipation, and loss of sense of smell, typically occur many years before the appearance of the PD motor symptoms that lead to a diagnosis. The loss of dopaminergic neurons in the substantia nigra, which leads to the motor symptoms seen in PD, is associated with the deposition of aggregated, misfolded α-Synuclein (α-Syn, *SNCA*) proteins forming Lewy Bodies. Additionally, dysregulation of miRNA (a short form of mRNA) may contribute to the developing pathophysiology in PD and other diseases such as cancer. Overexpression of α-Syn and miRNA in human samples has been found in PD, AD, and dementia. Therefore, evaluating these molecules in urine, present either in the free form or in association with extracellular vesicles of biological fluids, may lead to early biomarkers for clinical diagnosis. Collection of urine is non-invasive and thus beneficial, particularly in geriatric populations, for biomarker analysis. Considering the expression and function of α-Syn and miRNA, we predict that they can be used as early biomarkers in the diagnosis and prognosis of neurodegenerative diseases.

Keywords Parkinson's disease · Motor symptoms · Non-motor symptoms · Lewy body · miRNA · Urine

Introduction

Parkinson's disease (PD), sometimes referred to as "paralysis agitans," is an idiopathic and genetic disease of the nervous system characterized by motor and non-motor dysfunctions. This is a chronic, progressive neurodegenerative disease that appears predominantly in persons over the age of sixty while remaining rare among people under the age of forty (Kempster et al. 2010). PD is the second most common neurodegenerative disease behind Alzheimer's disease (AD), affecting ten million people worldwide. A record one million Americans are currently suffering from PD, with approximately 60,000 new cases occurring each year (Chou 2016; Parkinson's Disease Foundation: Statistics on Parkinson's 2016; Sherer et al. 2012). Many symptoms are debilitating, including visual hallucinations, fluctuations in consciousness, sleep disturbances, falls, and depression, all leading to loss of independence, disability, and a significant burden for caregivers.

Environmental and genetic factors can both contribute to the prevalence of PD. A genetic association is minor, as most cases are idiopathic; however, it has been well established that

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constitutive translation of the *SNCA* gene mediated by a 5′ untranslated region (5'-UTR) has an integral role in PD pathology (Koukouraki and Doxakis 2016). The protein, α-Synuclein (α-Syn), accumulates to form toxic, pathological forms such as α-Syn oligomers (Atik et al. 2016). Thus, recent studies on α-Syn oligomers and other toxic species of α-Syn (such as A53T, H50Q mutations) have led to the emergence of potential biomarkers in brain diseases (Kalia et al. 2013; Kasten and Klein 2013; Shalash et al. 2017). Another promising biomarker, miRNAs, are short regulatory RNAs that control gene expression through interacting with the UTR of target messenger RNA, causing translational repression or RNA destabilization (Filipowicz et al. 2008; Filipowicz et al. 2005). miRNA as a signature novel biomarker has been studied in colorectal cancer, Crohn's disease, and cardiomyopathy (Liu et al. 2013; Luo et al. 2011; Miyamoto et al. 2015; Zahm et al. 2011). Moreover, miRNA such as miR-135b-3p, miR-30e-5p, miR-223-3p, which have inflammatory pathway targets, are dysregulated in the blood, brain, and other body fluids in a broad range of diseases in adults, including neurodegenerative diseases PD, AD, and dementia (Cheng et al. 2015; Mancuso et al. 2019; Zeng et al. 2019).

The immune cells of the brain, microglia, have a considerable role in neurodegenerative diseases. They surveil the brain and are activated due to neuronal damage, misfolded proteins, viruses, and more. Microglial activation causes cytokine release and thus neuroinflammation, which, when chronic, results in cell death, as shown in Fig. 1 (Ouchi et al. 2009). It is still controversial as to whether neuroinflammation is a cause or a result of PD. Moreover, misfolded, oligomeric, and other unusual forms of α-Syn activate microglia; Increased microglia were found in the area of the brain most affected in PD, the Substantia Nigra (Lim et al.) of PD postmortem brain sections (Wakade et al. 2014). This suggested the infiltration of immune cells crossing the blood-brain barrier (BBB) (Banati et al. 1998; McGeer et al. 1988). Microglia can release anti-inflammatory cytokines and rescue injured cells, or they can take up dead or damaged cells, utilizing both autophagy and exosomes for clearance (Zhao and Wang 2019). The over-the-counter vitamin, niacin, was found to induce macrophage polarization and anti-inflammatory cytokines in PD (Wakade et al. 2014; Wakade et al. 2018). An in vitro study showed the ameliorative effect of niacin in regulating inflammatory cytokine through G protein-coupled receptor GPR109A via nuclear factor-kappa B (NF-kB) (Giri et al. 2019; Seamon et al. 2020). Reducing neuroinflammation is an important target for slowing or halting PD progression. The appearance of signature molecular events that can be seen in the body fluids, such as with neuroinflammation, could be observed much earlier than PD's clinical symptoms (Fig. 2).

Here, in this review, we discuss important considerations for urine as a biospecimen to identify early biomarkers. To provide further background information and justification for detecting the above molecules, we discuss the scope of available methods and instruments available for urinary analysis. We further present numerous urine assays currently used clinically, including high throughput methods and their importance to healthcare and clinical diagnosis, with a particular focus on microfluidic urine assays. Together, this review discusses the prospective candidate biomarkers, α-Syn, and miRNA, particularly in urine (a non-invasive method) which may have the ability to detect crucial warning signs, allow for earlier intervention and care, or reduce the risk of permanent or life-threatening damage of chronic neurodegenerative diseases.

Urine as a source of biomarkers

Early biomarker identification using liquid biopsy is key for timely, earlier diagnosis and to find preventative measures for the progression of PD, further neuronal loss, dementia, and disability. Moreover, non-invasive biospecimen collection methods are more attractive in geriatric subjects than healthy adult subjects due to the availability of healthy, accessible veins. Thus, urinary analysis has great potential in this respect, considering its biological richness and its capacity to be a convenient and cost-effective method for disease diagnosis/prognosis. A large volume of urine is produced daily by the average person: 6–7 urinations totaling 400 to 2000 mL; therefore, samples are abundant for collection and analysis (Lepowsky et al. 2017). It has been reported that urine contains more than 3000 biological molecules. A complete set of 2651 confirmed human metabolites and their structure has been documented (Bouatra et al. 2013). Even chronic diseases like cancer and coronary heart diseases use urine as a means of specific disease identifications (Brindle et al. 2002; Chan et al. 2011). Urine is also commonly used for clinical assessment in urinary tract infections (UTI), kidney function, diabetes, and pregnancy testing, but not in the context of brain diseases. This may be due to urine being conventionally regarded as an inappropriate source of biospecimens for neurodegenerative diseases since it is anatomically distant from the brain. The circulatory α-Syn oligomer and miRNA have no boundary (Table 1 & 2) as evidenced by the literature review. Several studies have indicated the overexpression of key proteins including α-Syn and miRNA, in biofluids of PD patients (Atik et al. 2016). These two molecules are promising potential candidates for early biomarkers in urine, allowing for screening and diagnosis that would enable the patients to receive proper care for brain diseases promptly (Simerville et al. 2005). Particularly for PD, early intervention enabled by early diagnosis could reduce the risk of neurological complications, motor function failure, or even death, all of which are associated with chronic neurodegenerative diseases.

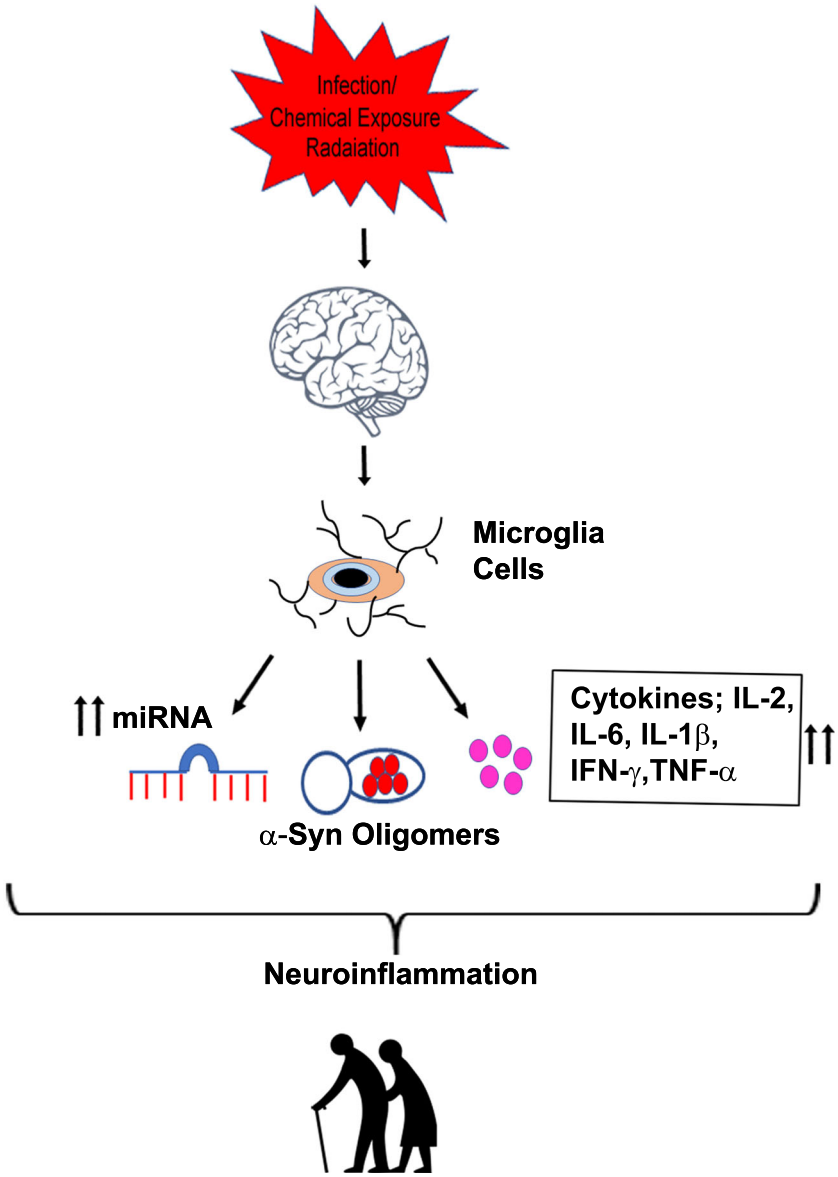

Fig. 1 Disease Pathway and emerging potential biomarkers. We postulate that stimulation of microglia cells through infection, chemical exposure, or radiation causes upregulation of α-Syn oligomers, miRNAs, and pro-inflammatory substances which ultimately mediate neuroinflammation in Parkinson's disease

α-Synuclein as a potential biomarker in PD

PD is considered a synucleinopathy that develops slowly; however, there is no efficient early diagnosis method, nor is there a cure. Mainly three different types of synucleinopathies exist, PD, dementia with Lewy bodies (DLB), and Multiple System Atrophy (Ng et al. 2011), which are characterized by the aberrant accumulation of the presynaptic protein α-Syn (*SNCA*), resulting in toxic α-Syn oligomers or Lewy Bodies (Ioanna Chalatsa et al. 2020). Different types of biofluids considered for the identification of candidate biomarkers such as blood, CSF, urine, and saliva have been summarized in Table 1 (Cao et al. 2019; Chang et al. 2019; Chen et al. 2020; Devic et al. 2011; Ioanna Chalatsa et al. 2020; Lee et al. 2006; Lin et al. 2017; Mollenhauer et al. 2019b; Mollenhauer et al. 2011; Nam et al. 2020; Pchelina et al. 2011; Shahnawaz et al. 2017; Tokuda et al. 2010; Vivacqua et al. 2019; Wang et al. 2015; Waragai et al. 2010). Several studies have reported evaluation of α-Syn in blood and CSF using commercially available Enzyme-linked immunosorbent assays (Psihogios et al.) kits and semi-quantitative immunoblot analysis in PD subjects (Lee et al. 2006; Waragai et al. 2010). On the contrary, some investigators failed to detect α-Syn in CSF (Jakowec et al. 1998; Ohrfelt et al. 2009). This suggests α-Syn is not a good candidate biomarker in CSF samples. Moreover, CSF sample collection is a cumbersome and painful method for geriatric subjects, particularly PD subjects with motor and non-motor dysfunction. A larger multicenter trial (MCT) may be needed to obtain specificity and sensitivity to detect α-Syn protein in CSF. However, plasma samples may offer better opportunities to identify α-Syn as a biomarker for PD. In this context, Lee et al. were able to

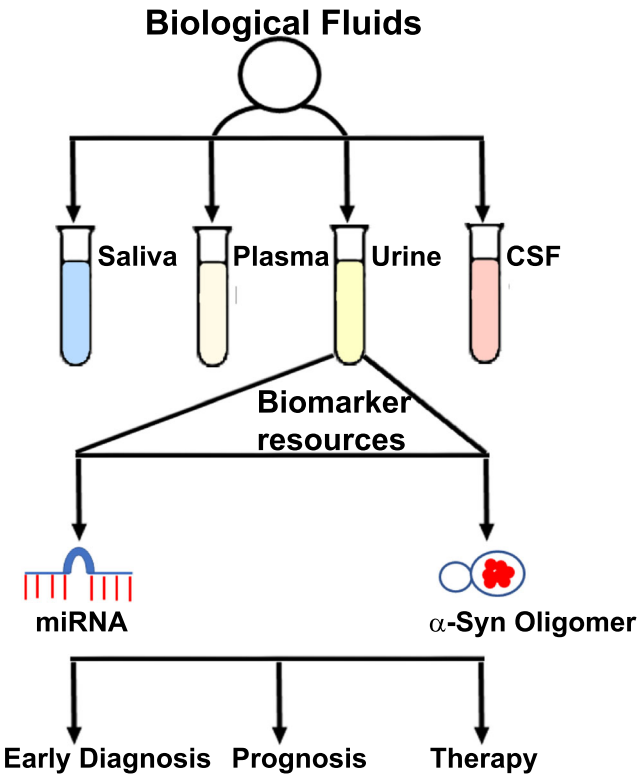

Fig. 2 Schematic representation of the biomarker resources for clinical diagnosis. The miRNAs and α-Syn oligomers, which are elevated in biological fluids such as saliva, plasma, urine, and CSF, could provide useful biomarkers for early diagnosis, prognosis, and therapeutic invention of Parkinson's disease

identify α-Syn in the blood of PD and MSA patients (Lee et al. 2006). The distribution of α-Syn in various body fluids and methods of detection has been given in Table 1. Several factors may account for the discrepancy in the results, including variations in specificity, sensitivity, and accuracy of each experimental method. This raises the question of how many samples were used in the assays and assay format capabilities for clinical diagnosis. Therefore, the concentration of analytes in clinical specimens is essential for evaluating molecules of interest.

We also believe, based on literature reviews by others, that there is a considerable variation in the concentrations of metabolites in urine, mainly due to the influence of many factors including age, gender, genetic background, health status, activity level, and diet (Guneral and Bachmann 1994; Psihogios et al. 2008; Zuppi et al. 1997). However, urine is a notable non-invasive biospecimen that could be available in a copious amount with no limitations for clinical diagnosis. α-Syn is constitutively expressed in various cells and transported across all peripheral tissue and body fluids (Shin et al. 2000). Recent studies showed α-Syn to be associated with many pathways, including endocytosis, Golgi homeostasis, ER-to-Golgi transport, and presynaptic trafficking for intercellular communications through exosomes (Loov et al. 2016; Wang and Hay 2015). Moreover, it exists in different forms, i.e., monomeric, oligomeric, and phosphorylated forms in different cells and the extracellular matrix (Surguchov 2016). The ubiquitous presence of α-Syn may offer an opportunity to evaluate its presence in non-invasive samples for clinical diagnosis.

Recently, several studies reported α-Syn was contained in and secreted through exosomes as a cargo vehicle to distant

Table 1 Historic data on α-Synuclein identifications in body fluids by different platforms

Detection/Changes in α-Synuclein	Disease State	Body Fluid	Methods	References
α-Syn	ND	CSF	ELISA	Ioanna et al. 2020
α-Syn	PD	CSF	ELISA	Mollenhauer et al. 2019
Oligomers ↑	PD	CSF	ELISA	Tokuda et al.
misfolded aggregates	PD	CSF	PMCA	Shahnawaz et al. 2017
Autoantibodies ↑	PD	Serum	ELISA	Shalash et al. 2017
α-Syn		Plasma & Serum	IMR Assay	Chang et al. 2019
α-Syn	PD	Plasma	IMR Assay	Lee et al. 2006
α-Syn	PD	Plasma	IMR Assay	Chen et al. 2020
Oligomers	PD	RBC	ELISA	Wang et al. 2015
α-Syn ↓	PD	Leukocytes	WB	Pchelina et al. 2011
α-Syn	PD & PSP	Saliva	ELISA	Vivacqua et al. 2019
α-Syn	PD	Saliva EVs	ELISA	Cao et al. 2019
α-Syn	PD	Saliva	WB	Devic et al. 2011
Oligomers	PD	Urine	ELISA	Nam et al. 2020

Abbreviations: ND – Neurodegenerative Disease, PD – Parkinson's Disease, PSP – Progressive Supranuclear Palsy, CSF- Cerebrospinal Fluid, RBC-Red Blood Cells, EVs – Extracellular Vesicles, ELISA- Enzyme-Linked Immunosorbent Assay, WB-Western Blot, IMR-Immunomagnetic Reduction Assay

Table 2 Common predictive miRNAs in the regulation of targeted genes in neurodegenerative diseases

Disease	miRNAs	Sample Type	mRNA Targets	References
Alzheimer's Disease	miR-107 ↓	Brain	BACE1	Wang et al. 2008b
	miR-30e-5p ↑	Exosomes	SOCS1, SOCS3	Cheng et al. 2015
	miR-485-3p↓	Brain & Serum	AKT3	Lau et al. 2013; Yu et al. 2020
	miR-200c ↑	Plasma	PTEN	Wu et al. 2016
	miR-210 ↓	CSF & Serum	VEGF	Zhu et al. 2015
	miR-34c ↑	Brain & Serum	SIRT1	Muller et al. 2014
Parkinson's Disease	miR-133b ↑	Brain	Pitx3	Kim et al. 2007
	miR-433 ↓	Serum & CSF	FGF20, JAK2	Wang et al. 2008a; Wang and Zhang 2020b
	miR-703		α-Syn	Nelson et al. 2008
	miR-148a,b		α-Syn	Nelson et al. 2008
	miR-153 ↓	CSF exosomes	a-Syn	Mouradian 2012
	miR-7		a-Syn, Nrf2, VDAC1	Mouradian 2012
	miR-223-3p ↑	Serum	NLRP3	Mancuso et al. 2019
	miR-125	Exosomes	α-Syn	Wang and Zhang 2020a
	miR-210	Exosomes	α-Syn	Wang and Zhang 2020a
	miR-450b	Exosomes	α-Syn	Wang and Zhang 2020a
	miR-669b	Exosomes	α-Syn	Wang and Zhang 2020a
	miR-494-3p	MPTP Mouse brain	SIRT3	Geng et al. 2018
	miR-135b-3p ↓	MPP+ Cells	NLRP3, FoxO1	Zeng et al. 2019
Multiple System Atrophy (Ng et al.)	miR-451 ↑	Serum	TSC1	Kume et al. 2018
	miR-101 ↑	Brain	ATG4D, Rab5A, mTOR	Valera et al. 2017
	miR-433 ↓	Brain	HDAC6	Lee et al. 2015
Dementia with Lewy Bodies	miR-9	Brain	MYO1D, PSEN1	Pietrzak et al. 2016
	miR-124	Brain	MAP7	Pietrzak et al. 2016
Huntington Disease	miR-132 ↓	Brain	P250GAP	Scott et al. 2012; Johnson et al. 2008
	miR-520f-3p ↑	CSF	SOX9	Reed et al. 2018
	miR-30a	Brain	REST, BDNF	Hwang and Zukin 2018; Marti et al. 2010
	miR-342-3p		HIP1	Bhattacharyya et al. 2008
Prion disease	miR-191 ↑	Brain	EGR1	Saba et al. 2008
	miR-342-3p		MSI1, TTBK2	Montag et al. 2009
	miR-494 ↑	Macaque brain	ATRX, SFRS7, RDJ2	Beck et al. 2006

Abbreviations: BACE1- Beta Secretase 1; SOCS – Suppressor of cytokine signaling; AKT3 - RAC-gamma serine/threonine-protein kinase; PTEN - Phosphatase and tensin homolog; VEGF – vascular endothelial growth factor; SIRT - NAD-dependent deacetylase sirtuin; Pitx3 - paired-like homeodomain transcription factor 3; FGF20 - Fibroblast growth factor 20; JAK2 – Janus Kinase 2; α-Syn - Alpha-Synuclein; Nrf2 - Nuclear factor erythroid 2-related factor 2; VDAC1 - Voltage-dependent anion-selective channel 1; NLRP3 - NOD-, LRR-, and pyrin domain-containing protein 3; FoxO1 - Forkhead box protein O1; TSC1 - Tuberous sclerosis protein 1; ATG4D - Autophagy related 4D, cysteine peptidase; Rab5A - Ras-related protein 5A; mTOR **- Mechanistic target of rapamycin;** HDAC6 - Histone deacetylase 6; MYO1D – myosin ID; PSEN1 – Presenilin-1; MAP7 – microtubule-associated protein 7; P250GAP - GTPase-activating protein 250; SOX9 - SRY-Box Transcription Factor 9; REST-RE1 Silencing Transcription Factor; Chk1-Checkpoint kinase 1; BDNF - brain-derived neurotrophic factor; HIP1 -Huntingtin-interacting protein 1; EGR1 - Early growth response protein 1; MSI1 - musashi1, an RNA-binding protein; TTBK2-Tau tubulin kinase 2; ATRX-thalassemia/mental retardation, X-linked; SFRS7 - Splicing Factor, Arginine/Serine Rich 7; RDJ2 - a J protein family member, interacts with cellular prion PrP(C)

tissues and/or for communication and excretion through the skin, saliva, blood, and CSF (Mollenhauer et al. 2019a; Mollenhauer et al. 2019b; Mollenhauer et al. 2019c). Ho, et al. showed increased levels of DJ-1 and LRRK2 proteins detected in the exosomes isolated from urine samples from PD patients, but they were not able to detect α-Syn in the urine exosomes (Ho et al. 2014). Ioanna et al. found that Lewy Bodies in PD pathology are mostly made up of α-Syn that has been excessively phosphorylated, proposing phosphorylated α-Syn as another potential early biomarker (Ioanna Chalatsa et al. 2020). The precise and site-specific measurements of α-Syn expression levels may be relevant to disease specificity and are necessary for detecting the quality and quantity of this protein for clinical diagnosis. The existence of the differing forms of α-Syn may reflect specific disease activities, and the availability of urine could be obtained easily from any patients, including PD subjects. Identifying and quantifying the different forms of α-Syn in urine may be a novel method to discriminate brain diseases from other unrelated neurological disorders for clinical diagnosis and to implement early intervention measures for delayed progression of the disease.

α-Synuclein determination in urine samples

Urine was conventionally regarded as an inappropriate source of biomarkers for neurodegenerative diseases because it is anatomically distant from the brain compared to peripheral blood and CSF (Reed et al.). However, recent studies have suggested that urine could be utilized as a source of brain disease biomarkers if an appropriate marker is predetermined by immunoassay approaches in PD subjects' urine (Nam et al. 2020). Studies have also demonstrated an accumulation of α-Syn in the urine of animals and humans under both physiological and pathological states (Seol et al. 2020). A few urinary biomarkers already found such as formaldehyde and 8-OHdG, show a correlation with severity of the disease (Nam et al. 2020; Seol et al. 2020). There are limited studies using urine to evaluate α-Syn as a biomarker.

Next-generation technologies will lead to developing affordable and accessible healthcare. However, the advent of recent sophisticated technology includes proteomics and paper-based detection systems, which may be applied in the identification and analysis of α-Syn and alterations of protein levels in urine relative to the severity of the disease. Mass spectrometry provides the best sensitivity, selectivity, and quantitative identification capabilities to determine the bioactive compounds with accuracy (Beasley-Green 2016). (Psihogios et al.) already tested and validated by Nam et al. also have the sensitivity to quantitatively detect α-Syn oligomers in urine in PD subjects compared to age-matched control groups (Table 1) (Nam et al. 2020). Moving forward in the development of paper-based microfluidic urine analysis would be beneficial for promoting efficient, effective, and affordable diagnosis. In this context, Lepowsky et al. described the paper-based urine analysis methods as a point of care diagnosis (Lepowsky et al. 2017). It is predicted that α-Syn oligomers could be identified using paper-based technology with more sensitivity and specificity than classical immunoassays and immunoblots for clinical diagnosis and prognosis of PD and other synucleopathic diseases.

Circulating miRNAs as potential biomarkers for PD

miRNAs regulate gene expression at the post-transcriptional level has been well established. They have recently emerged as prominent transcriptional regulators of various cellular processes, including neurodevelopment. They are abundantly present in the brain, and their dysfunction has been implicated in an array of neuropathological conditions, including PD, AD, and dementia. miRNAs exist in the tissues and have also been found to circulate in several body fluids, including plasma or serum, CSF, urine, and saliva. There are significant differences between the circulating miRNA expression profiles of healthy individuals and those of PD patients. The importance of miRNA in biological processes has been recently recognized for its potential role in regulating approximately 33% of human genes (Ambros 2004; Lewis et al. 2005; Lim et al. 2003). Changes in miRNA expression have been associated with the development and progression of diseases (Tili et al. 2007). Recent studies evaluated miRNAs as a potential for diagnostics in many clinical applications, including cancer. However, there are limitations of miRNA-based diagnostics in brain diseases due to the impracticality of sampling brain tissues from living humans. Difficulties also occur with obtaining CSF and blood samples from aged populations. Growing evidence based on several studies revealed miRNA to be a novel class of biomarkers for disease diagnosis and prognosis in PD (Du et al. 2009; Guerau-de-Arellano et al. 2012; Otaegui et al. 2009). There are two notable reasons why miRNA should be considered as potential biomarkers for disease identification: the expression level of miRNAs that are specific to the disease's activities and the availability of miRNAs in a large number of samples, including tissue biopsies, whole blood, blood cells, serum, plasma, urine, and even exosomes.

Urine is considered one of the prominent extracellular human body fluids obtained in large volumes using simple, non-invasive methods and is easy to obtain in large volumes for clinical diagnosis and disease identifications. Even chronic diseases like cancer and coronary heart diseases use urine as a means of specific disease identifications (Brindle et al. 2002; Chan et al. 2011). Recently, several miRNAs have been detected in urine samples (Hanke et al. 2010). Several supporting articles identify more than 40 urinary miRNAs related to specific urological pathologies, including intrinsic kidney disease, renal cell carcinoma, and bladder cancer (Sun and Lerman 2019; Thorsen et al. 2012; Zahm et al. 2011). There is a significant concern about the stability of miRNA in the harsh urinary milieu; However, the miRNA can also be tightly packed in exosomes which are protected from the harsh conditions of urine (Street et al. 2017). The exosome not only provides protection but also keeps miRNA in a concentrated form, making robust detection quantitative for disease identifications with accuracy.

Table 2 shows the expression of disease-specific miRNA profiles targeting proteins of interest unique to various neurological diseases. Specifically, several miRNAs, such as miR-21, miR-192, miR-200, and miR-29, are upregulated in renal diseases described by Chung et al. (Chung et al. 2013; Sun and Lerman 2019). Moreover, miRNAs profiles are not limited to renal diseases and are also found in the blood, CSF, saliva, and urine in response to central nervous system injury (Sun et al. 2018). Several miRNAs (e.g., miR-497, Let-7f, miR-181, miR-15a/16–1, miR-23a, miR-424, miR-124, miR-122, miR-21, others) that are altered after CNS injuries have been reported, relating to neuroinflammation in nervous

systems (Sun et al. 2018). Microarray would be ideal for capturing miRNA changes related to disease type and severity (Jimenez-Avalos et al. 2020). miRNAs are not only used for diagnostic parameters, but it has a potential role in gene therapy for regulating disease-modifying genes of interest (Hashemi and Gorji-Bahri 2020; Thorsen et al. 2012; Zhao and Wang 2019). With the development of the latest innovative technology, including paper-based microfluidic technology, LC-MS/MS, inductively coupled plasma mass spectrometry, and high-performance liquid chromatography (HPLC), numerous metabolic molecules can be determined both qualitatively and quantitatively with accuracy in very short periods of time (Bouatra et al. 2013).

Despite the challenges, a miRNA-based diagnosis has become a promising strategy for neurodegenerative disease identifications either at point-of-care diagnosis or with sophisticated instruments in laboratory settings. Much remains to be discovered in order to utilize miRNA-based approaches and identify candidate miRNAs as potential biomarkers in brain diseases. Successful clinical identification and validations of miRNAs in urine will undoubtedly enhance the future of this nascent field.

Conclusion

In general, PD itself does not cause death but is associated with an increased risk of morbidity and mortality worldwide (Hirsch et al. 2003; Lindqvist et al. 2012). Despite the numerous research studies, clinical diagnosis and management of PD are still in their infancy by suboptimal detection and prognosis methods. There are no optimized, validated biomarkers available with high enough sensitivity and specificity for early disease identification before the canonical PD motor symptoms occur. Early diagnosis of the disease is essential for enhanced maintenance of PD subjects' health through clinical interventions. Here we highlighted both α-Syn oligomers and miRNAs as potential biomarkers for disease diagnosis and explored the use of urine as a potential non-invasive biospecimen sample to identify biomarkers for neurodegenerative diseases, specifically PD. Recent technological advances allow for the systematic, holistic, and unbiased identifications and characterization of miRNAs and α-Syn oligomers in the urine, ensuring the promising potential of approaches in biomarker discovery. Developing optimal miRNA and α-Syn oligomer identification tools for clinical validation may pave the way for novel biomarkers in PD diagnosis and prognosis.

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Code availability Not Applicable.

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Draft, BG, MS, AB.
Revisions, BG, MS, AB, SC, MCW, JM, SP, BB.

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Data availability Not Applicable.

Declarations

Ethics approval This article does not contain any studies with human participants or animals performed by any of the authors.

Conflict of interests The authors declare no conflict of interest.

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