Precision medicine to identify optimal diagnostic and therapeutic interventions for Parkinson's Disease

Tarek Elshourbagy[1,2]*, Alveena Batool Syed[3], Mohamed A.M. Amer[4], James Robert Brasic[5]

1 Faculty of Medicine, Cairo University, Egypt
2 Rutgers New Jersey Medical School, Newark , NJ, USA
3 Liaquat University of Medical and Health Sciences, Jamshoro, Pakistan
4 Faculty of Medicine, Menoufia University, Egypt
5 The Johns Hopkins University School of Medicine, Section of High Resolution Brain Positron Emission
  Tomography Imaging, Division of Nuclear Medicine and Molecular Imaging, Baltimore, MD 21287, USA

* **Corresponding Author:** Tarek Elshourbagy **E-mail:** doctarek_medico@yahoo.com

ABSTRACT

Objective: Parkinson's disease, the second most common neurodegenerative disorder afflicting 10 million people worldwide and the fourteenth leading cause of death in the United States, is caused by the death of dopaminergic neurons that regulate movement in the substantia nigra pars compacta. Mechanisms contributing to the development of Parkinson's disease in vulnerable individuals include protein misfolding, protein aggregation, and mitochondrial dysfunction. In order to develop guidelines for clinicians to utilize precision medicine to develop treatment plans to address the specific needs of individuals with Parkinson's disease and related conditions, we have developed algorithms for diagnosis and treatment based on their view of available knowledge. We reviewed the key literature on the pathogenesis of Parkinson's disease on PubMed and google scholar in order to propose guidelines for the development of diagnostic and therapeutic interventions for people with Parkinson's disease and related conditions. In about 25 percent of patients, clinicians incorrectly diagnose Parkinson's disease. Causes of misdiagnosis include a lack of algorithms and inadequate use of diagnostic modalities. Four main mechanisms that may contribute to the development of Parkinson's disease (misfolding of alpha-synuclein, mitochondrial dysfunction, dysfunctional ubiquitin proteasomal pathways, and abnormal autophagy) and different diagnostic modalities (structured interview and examination, laboratory assessments, neuropathology, genetic testing, neuroimaging) will form the basis for our algorithm for the diagnosis and treatment of Parkinson's disease and related conditions. Clinicians, administrators, policy planners, advocates, and other concerned individuals will benefit from the adoption of our guidelines for the diagnosis and treatment of Parkinson's disease and related conditions.

Keywords: Precision Medicine, Parkinson's Disease, Algorithms, Basal ganglia, Neurodegenerative disorders, DAT-SPECT, L-DOPA, Dopamine agonists, Amyloid Protein, Proteosomes, Neuroimaging, Accelerometer, CT, MRI, PET, Transcranial Sonograghy, Neurosurgery, Pallidotomy, Thalamotomy, Sargramostism, Coenzyme Q10, Genetic Testing, Polymerase Chain reaction, Ubiquitination Assay, PRKN gene

INTRODUCTION

Pathogenesis of the Parkinson's Disease

a) Alpha-synuclein protein misfolding

Alpha-synuclein is a cerebral protein normally found in an insoluble fibril. It is an abundant presynaptic protein that binds to negatively charged phospholipids, which also function as SNARE –complex chaperone which contributes to the pathogenesis of Parkinson disease due to its misfolding (1).Experiments were carried out on rodents and found the abnormal neurotoxic alpha-synuclein protein is found in an oligomeric form rather than the mature insoluble fibrils (2). Protein folding and refolding of the misfolded proteins occurs via a group of molecules known as chaperones and co-chaperones such as Hsp70 and Hsp90 and their Co Chaperone Hsp 40 (3). Syncluein misfolding can be detected via a molecular test called Protein Misfolding Cyclic Amplification that will be discussed in detail (4).

Developing drugs that enhance chaperone and co-chaperone molecular systems may help reduce protein misfolding in some patients with Parkinson's disease (3).

### b) Dysfunctional ubiquitin-proteasome system

Proteosomes are organelles responsible for protein turnover. Diminished activity of these organelles may result in abnormal protein accumulation in the basal ganglia and cause damage which may contribute to the development of Parkinson's disease (5). Decreased expression of proteasomal components has been identified in the SNpc of the brain in patients with Parkinson's disease (6). Ubiquitination Assay for Parkin gene can be tested in autosomal recessive cases of PD (7).

c) Mitochondrial dysfunction

Mitochondrial dysfunction has been found in many cases of idiopathic or hereditary Parkinson's disease (8).The neuro-toxic alpha-synuclein in its oligomeric form can interact with a mitochondrial receptor resulting in increased oxidative damage due to reduced mitochondrial respiration (9).Genetic testing can help diagnose hereditary cases with mitochondrial dysfunction as many genes involved have been detected as SNCA gene mutation in Autosomal dominant cases while Parkin gene mutation in Autosomal recessive cases. Developing drugs that target Mitochondrial Complexes to resolve the dysfunction may enhance symptoms in PD (10).

Diagnostic Modalities

a) The most recent simple diagnostic modality: Tri-axial accelerometer (11).
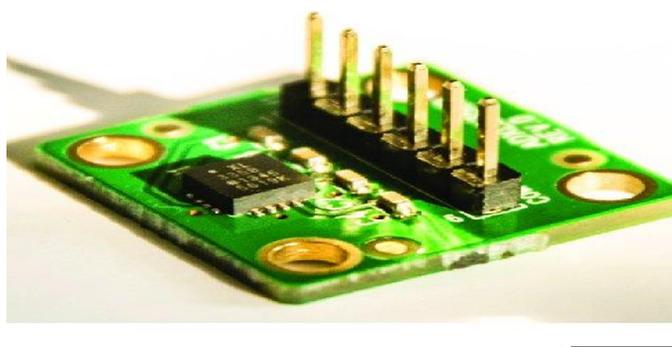
McKay, James Brasic, and their colleagues at Johns Hopkins medical school developed a low cost simple objective method to diagnose and grade Parkinson's disease (11). This technique involves objective quantitative assessment of tremors in extremities of patients with Parkinson's disease.

Low-cost accelerometers are attached to the upper and lower limbs to generate a continuous three diminutions representation of the movements. Patients and controls were assessed through a test-retest method. The degree of tremors is rated clinically by trained examiners. The output of the accelerometers can be interpreted using Fast Fourier Transforms and Continuous Wavelet transforms by experts for diagnosis and therapy (12). The use of this device for clinical practice is not FDA approved yet (11).

b) Transcranial sonography

Sonography is a simple, low-cost and widely available imaging modality and it can be used for diagnosing Parkinson's disease with high accuracy. Pooled sensitivity and specificity of transcranial -sonography for diagnosing Parkinson's disease is 84% and 85%. This is calculated from the meta-analysis of 39 studies involving 3123 patients (13). Usually, it is not used for diagnosis unless there is an associated condition such as epilepsy or stroke.

c) Genetic and molecular testing

1) RT-Polymerase chain reaction (RT-PCR)

This method can detect genetic mutations in hereditary cases of PD. It can be used for screening and diagnosis. This method uses heat to denature DNA then annealing occurs via Reverse Transcription DNA polymerase enzyme to amplify the gene of interest (14).

2) Protein Misfolding Cyclic Amplification

This test uses ultrasound waves to degrade proteins such as alpha-synucleininto small pieces so they can be amplified and studied. This method can be used to detect misfolded alpha-synuclein protein with high sensitivity and specificity in patients with multiple system atrophy and Parkinson's disease (15).

3) Ubiquitination assay for ligase enzyme

This method can be used in Autosomal recessive cases with Parkin gene mutations. The concept relies on detecting the activity of the Parkinubiquitin ligase enzyme which is important for protein turnover (16).

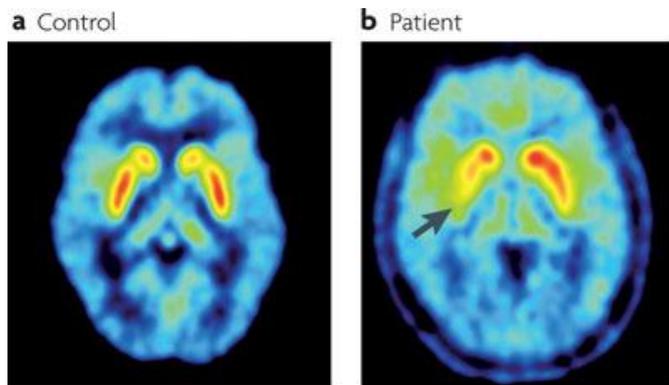### d) Molecular imaging methods

1) In single-photon emission computed tomography (SPECT) (33).
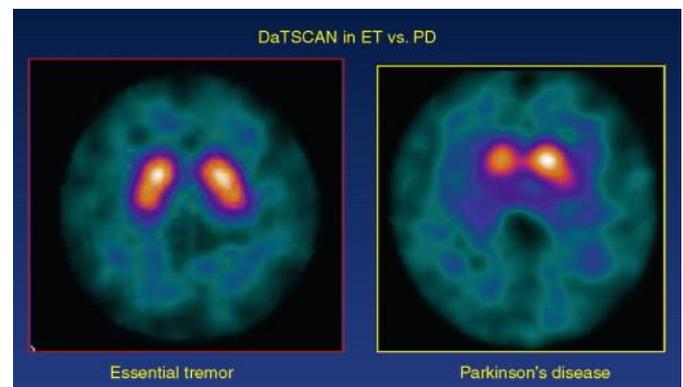
SPECT is a method of molecular imaging in which a gamma ray-emitting radioactive isotope is tagged to a molecule of interest. In patients with Parkinson's disease, labeled cocaine derivatives such as Ioflupane for DAT-SPECT is most widely used, others include 123I-β-CIT and 123I-FP-CIT (N-ω-fluoropropyl-2β-carboxymethoxy-3β-(4-iodophenyl tropane). They label the presynaptic dopamine reuptake sites. Patients with Parkinson's disease show reduced uptake of these substances in the basal ganglia (17). A subtype of SPECT called 123I-β-CIT SPECT was 100% sensitive and specific for the diagnosis in younger patients (age <55 years). In older patients (age >55 years), specificity was substantially lower (68.5%) (18).

A major disadvantage of SPECT, when compared to other methods of nuclear imaging such as PET, include the limited resolution of images for the visualization of basal ganglia in PD (19).

## 2) Positron emission tomography (34)

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PET is another method of molecular imaging in which a positron-emitting radioactive isotope is tagged to a molecule of interest. Fluorine is injected intravenously into patients with Parkinson's disease. Fluorine can be attached to either DOPA or deoxyglucose. Flurodopa F18 is taken by the presynaptic neurons in the basal ganglia particularly the caudate and putamen. However, fluorodeoxyglucose is taken by active cells to be trapped in the tissues during the imaging study. Reduced uptake of Flurodopa is seen in Parkinson's disease (20).

One study showed that the use of F-DOPA PET to discover the pathogenesis of Parkinson's disease showed that F-DOPA uptake correlated with increased bradykinesia and rigidity, not with tremors which show that the pathogenesis of tremors is different from the pathogenesis of bradykinesia and rigidity (21).

It can be used if the diagnosis is inconclusive by DAT-SPECT.A meta-analysis calculated the pooled sensitivity of PET in diagnosing Parkinson's disease as 88% and specificity as 92 % (22). A major disadvantage of PET imaging is the short half-lives of the radioactive isotopes which do not allow much time for imaging. Another major disadvantage is the high cost of a PET scan.

3) Magnetic resonance imaging (35).
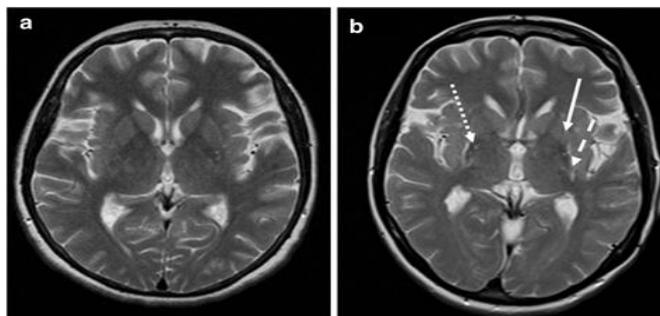
MRI is a method of imaging in which high magnetic field strengths are used to excite the hydrogen atoms in water molecules. MRI can demonstrate structural changes(atrophy) in the basal ganglia in patients with Parkinson's disease as illustrated in Image B.MRI is superior to CT, because of better resolution and sensitivity to identify structural brain pathology (23). A meta-analysis study published in 2021 calculated pooled sensitivity and specificity of using MRI in PD. The pooled sensitivity was found as 92% and pooled specificity as 90% (24). Usually, it is used for diagnosis only if there is an associated condition like stroke or epilepsy.

4) Magnetic resonance spectroscopy

MRS can be used to quantify the intermediary metabolites in small volumes of brain tissue. Patients with Parkinson's disease show decreased concentration of Dopamine in the basal ganglia. This method is usually used for research purposes (25).

Our Algorithm for Diagnosis: Algorithm for Diagnosis of PD (ADPD)

| Diagnosis of PD (ATPD)	
Step 1	**1. History of Present Illness:** History of Bradykinesia with/without static tremors and or rigidity. It is crucial to differentiate PD from other similar conditions such as • Supranuclear palsy which is characterized by gaze palsy seen early in the disease. • Shy-Dragersyndromewhichis characterized by autonomic dysfunction and • Cortical base degeneration which is characterized by early cortical signs as speech deficits and cognitive decline. **2. History of Traumatic brain injury if present.** **3. History of any chronic illness.** **4. Family history of PD and or other movement disorders.**
Step 2	**Clinical examination:** • Inspection: Static regular tremors like Bell rolling. • Tone:Rigidity of upper and or lower extremities. • Gait:Bradykinesia and characteristic shuffling gait.
Step 3	**Use of mechanical accelerometer.** • Simple and cheap method. • It allows accurate grading of tremors. If diagnosed: Step 4
Step 4	**With a family history.** • Genetic and molecular testing (RT-PCR,Ubiquitination Assay for ligase enzyme,etc) **Without Family history** a) If no other condition is associated • DAT SPECT. • Reliable and widely available Imaging Modality. b) If another condition associated (epilepsy or stroke) • Transcranial Sonography • CT • MRI
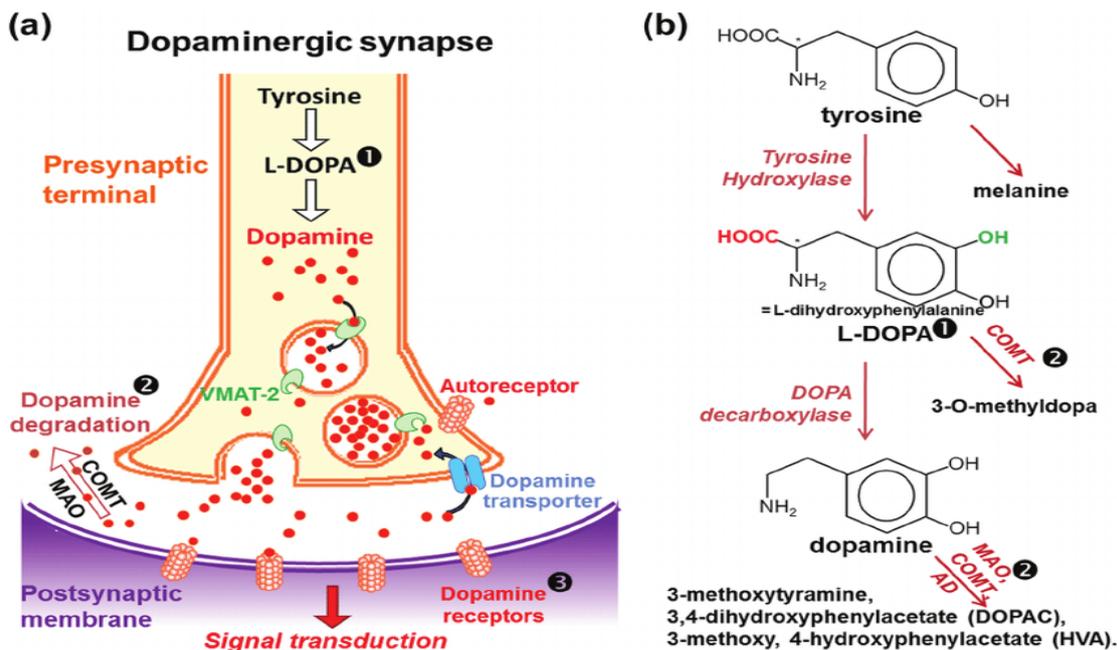
Figure: formation of Dopamine in the basal ganglia (36)

Medications used for Parkinson's disease:

Most medications used for Parkinson's disease target the neuro-transmitter; Dopamine which is formed in the basal ganglia in normal individuals.

To understand the mechanisms of these medications, we must discuss how Dopamine is formed normally in the basal ganglia.

Tyrosine is the amino acid used for the formation of Dopamine and other neurotransmitters as adrenaline and noradrenaline.

The picture below illustrates the steps for the formation of Dopamine in the basal ganglia

a) L-DOPA

L-DOPA is the most potent and is the first line of treatment for Parkinson's disease, especially in the early stages. It provides DOPA that is decarboxylated in the body to Dopamine. It is highly effective but it has many adverse effects. Combing it with Carbidopa helps prevents decarboxylation outside the brain which reduces adverse effects (26, 27).

b) Dopamine agonists

Drugs such as Pramipexole can be used to treat Parkinson's disease as they stimulate the dopaminergic 2 receptors which are located in the basal ganglia (27, 28).

c) Catechol methyl-transferase inhibitor

Tolcapone and Entacapone are drugs that work via inhibiting the enzyme Catechol Methyl-transferase thus increasing dopamine availability in the basal ganglia.

d) Monoamine oxidase inhibitors

Drugs that inhibit MAO –B such as Selegiline help increase the availability of Dopamine in the basal ganglia in a step different from COMT inhibitors. Both classes have similar efficacy (27, 28).

A meta-analysis summarized 45 clinical trials on 9000 patients to compare the efficacy of adjuvant drugs such as dopamine agonists, catechol-methyl-transferase inhibitors to L-DOPA. Dopamine agonists were found to be more effective than catechol-methyl-transferase inhibitors and MAO-B inhibitors which have similar efficacy. Drugs of the same class were not different in efficacy except Tolcapone which was found to be more effective than entacapone (29).

Experimental drugs

a) Carbenoxolone

Laboratory Experiments showed that the drug carbenoxolonein a low dose enhances the activity of Hsp 90 in rat models which results in the reduction of misfolded alpha-synucleinprotein in neurons and improvement in motor functions (29)

b) Coenzyme Q10(Ubiquinol-10)

Coenzyme Q10 serves as a co-factor for complexes 1,2 and 3 for the electron transport chain in the mitochondria. It can be of significant benefit for some patients with PD.A clinical study showed that it might be helpful in patients taking L-DOPA with wear-off symptoms (30).

c) Sargramostism(GM-CSF):

GM-CSF is a cytokine produced by T- regulatory cells and it plays a role in PD by protecting against basal ganglia degeneration (10, 30). Drugs targeting the immune system like Sargramostism has shown promise in animal model of Parkinson's disease and human clinical trials (31).

Surgical methods

a) Deep Brain Stimulation

The concept relies on stimulating the subthalamic nucleus which inhibits signals from the basal ganglia. It is an invasive method but showed marked improvement in many patients.

b) Thalamotomy

The concept relies on destruction of the thalamus. Benefit for tremors only.

c) Pallidotomy

The Concept relies on destruction of globus pallidum. Benefit for tremors, bradykinesia,and levodopa-induced dyskinesias. Bilateral procedures are not recommended.

d) Subthalamotomy

The concept relies on destruction of the subthalamicnucleas. Under development; may reduce tremor, rigidity, bradykinesia, and levodopa-induced dyskinesias. Not recommended for bilateral use.

Other surgical methods include focused ultrasound thalamotomies, and cell transplantation therapies (**32, 33**).

Our Algorithm for Treatment of PD: Algorithm for Treatment of PD (ATPD)

Treatment of PD (ATPD)	
Step 1	Exercise and rehabilitation medicine. L-DOPA or carbidopa. If no improvement(monitor degree of static tremors by accelerometer /monitor other signs of PD.
Step 2	Increase dose of L-DOPA/carbidopa. Maintain exercise andrehabilitationmedicine. If no improvement or wear-off symptoms (Monitor degree of static tremors by accelerometer /monitor other signs of PD.
Step 3	Maximize dose of L-DOPA/carbidopa with or without another class of medication such as MAO-B inhibitor or COMT inhibitor. Use of co-enzyme(Ubiquinol-10). Maintain exercise and rehabilitationmedicine. If no improvement, Monitor degree of static tremors by accelerometer /monitor other signs of PD.
Step 4	MAO-B inhibitors with or without COMT Inhibitors. Maintain exercise and rehabilitation medicine. If no improvement (monitor degree of static tremors by accelerometer /monitor other signs of PD.
Step 5	Deep brain stimulation (invasive method).
Step 6	Surgical methods including radiofrequency or radiosurgery procedures(pallidotomy,thalamotomy or subthalamotomy).
Step 7	Potential new treatments including immunotherapy, gene therapy or cell transplantation.

CONCLUSION

Clinicians, administrators, policy planners, advocates, and other concerned individuals will benefit from the adoption of our algorithms for the diagnosis and treatment of Parkinson's disease.

Author Contributions: TE, ABS, MAMA, JRB: Search of the literature, performed the analysis, wrote the manuscript, and designed the article for submission.

REFERENCES

1. Burré J, Vivona S, Diao J, Sharma M, Brunger AT, Südhof TC. Properties of native brain α-synuclein. Nature. 2013;498(7453):E4-6; discussion E6-7.

2. Winner B, Jappelli R, Maji SK, Desplats PA, Boyer L, Aigner S, et al. In vivo demonstration that alpha-synuclein oligomers are toxic. Proc Natl Acad Sci U S A. 2011;108(10):4194–9.

3. Jung BC, Lim Y-J, Bae E-J, Lee JS, Choi MS, Lee MK, et al. Amplification of distinct α-synuclein fibril conformers through protein misfolding cyclic amplification. Exp Mol Med. 2017;49(4):e314–e314.

4. Kalia SK, Kalia LV, McLean PJ. Molecular chaperones as rational drug targets for Parkinson's disease therapeutics. CNS Neurol Disord Drug Targets. 2010;9(6):741–53.

5. McKinnon C, Tabrizi SJ. The ubiquitin-proteasome system in neurodegeneration. Antioxid Redox Signal. 2014;21(17):2302–21.

6. McNaught KSP, Belizaire R, Jenner P, Olanow CW, Isacson O. Selective loss of 20S proteasome α-subunits in the substantia nigra pars compacta in Parkinson's disease. Neurosci Lett. 2002;326(3):155–8.

7. Schlossmacher MG, Shimura H. Parkinson's disease: assays for the ubiquitin ligase activity of neural Parkin. Methods Mol Biol. 2005;301:351–69.

8. Moon HE, Paek SH. Mitochondrial dysfunction in Parkinson's disease. Exp Neurobiol. 2015;24(2):103–16.

9. Di Maio R, Barrett PJ, Hoffman EK, Barrett CW, Zharikov A, Borah A, et al. α-Synuclein binds to TOM20 and inhibits mitochondrial protein import in Parkinson's disease. Sci Transl Med. 2016;8(342):342ra78-342ra78.

10. Thomas B, Beal MF. Mitochondrial therapies for Parkinson's disease: Mitochondria and Parkinson's Disease. Mov Disord. 2010;25(S1):S155–60.

11. McKay GN, Harrigan TP, Brašić JR. A low-cost quantitative continuous measurement of movements in the extremities of people with Parkinson's disease. MethodsX. 2019;6:169–89.

12. Brasic J. Signal processing of quantitative continuous measurement of movements in the extremities. Mendeley; 2020.

13. Tao A, Chen G, Deng Y, Xu R. Accuracy of transcranial sonography of the substantia nigra for detection of Parkinson's disease: A systematic review and meta-analysis. Ultrasound Med Biol. 2019;45(3):628–41.

14. Wang J, Liu Y, Chen T. Identification of key genes and pathways in Parkinson's disease through integrated analysis. Mol Med Rep. 2017;16(4):3769–76.

15. Shahnawaz M, Mukherjee A, Pritzkow S, Mendez N, Rabadia P, Liu X, et al. Discriminating α-synuclein strains in Parkinson's disease and multiple system atrophy. Nature. 2020;578(7794):273–7.

16. Benamer HTS, Oertel WH, Patterson J, Hadley DM, Pogarell O, Höffken H, et al. Prospective study of presynaptic dopaminergic imaging in patients with mild parkinsonism and tremor disorders: part 1. Baseline and 3-month observations: Presynaptic Dopaminergic Imaging. Mov Disord. 2003;18(9):977–84.

17. Eerola J, Tienari PJ, Kaakkola S, Nikkinen P, Launes J. How useful is [I123]β-CIT SPECT in clinical practice? Journal of Neurology, Neurosurgery and Psychiatry. 2005;76(9):1211–1216,.

18. Wang L, Zhang Q, Li H, Zhang H. SPECT molecular imaging in Parkinson's disease. J Biomed Biotechnol. 2012;2012:412486.

19. Vingerhoets FJ, Schulzer M, Calne DB, Snow BJ. Which clinical sign of Parkinson's disease best reflects the nigrostriatal lesion? Ann Neurol. 1997;41(1):58–64.

20. Otsuka M, Ichiya Y, Kuwabara Y, Hosokawa S, Sasaki M, Yoshida T, et al. Differences in the reduced 18F-Dopa uptakes of the caudate and the putamen in Parkinson's disease: correlations with the three main symptoms. J Neurol Sci. 1996;136(1–2):169–73.

21. Gu S-C, Ye Q, Yuan C-X. Metabolic pattern analysis of 18F-FDG PET as a marker for Parkinson's disease: a systematic review and meta-analysis. Rev Neurosci. 2019;30(7):743–56.

22. Brooks DJ. Morphological and functional imaging studies on the diagnosis and progression of Parkinson's disease. J Neurol. 2000;247 Suppl 2:II11-8.

23. Cho SJ, Bae YJ, Kim J-M, Kim HJ, Baik SH, Sunwoo L, et al. Iron-sensitive magnetic resonance imaging in Parkinson's disease: a systematic review and meta-analysis. J Neurol [Internet]. 2021; Available from: http://dx.doi.org/10.1007/s00415-021-10582-x

24. Clarke CE, Lowry M. Systematic review of proton magnetic resonance spectroscopy of the striatum in parkinsonian syndromes. Eur J Neurol. 2001;8(6):573–7.

25. LeWitt PA. Levodopa therapy for Parkinson's disease: Pharmacokinetics and pharmacodynamics: Levodopa Pharmacokinetics and Pharmacodynamics. Mov Disord. 2015;30(1):64–72.

26. Radhakrishnan DM, Goyal V. Parkinson's disease: A review. Neurol India. 2018;66(Supplement):S26–35.

27. Armstrong MJ, Okun MS. Diagnosis and treatment of Parkinson disease: A review: A review. JAMA. 2020;323(6):548–60.

28. Thakur P, Nehru B. Long-term heat shock proteins (HSPs) induction by carbenoxolone improves hallmark features of Parkinson's disease in a rotenone-based model. Neuropharmacology. 2014;79:190–200.

29. Yoritaka A, Kawajiri S, Yamamoto Y, Nakahara T, Ando M, Hashimoto K, et al. Randomized, double-blind, placebo-controlled pilot trial of reduced coenzyme Q10 for Parkinson's disease. Parkinsonism Relat Disord. 2015;21(8):911–6.

30. Gendelman HE, Zhang Y, Santamaria P, Olson KE, Schutt CR, Bhatti D, et al. Evaluation of the safety and immunomodulatory effects of sargramostim in a randomized, double-blind phase 1 clinical Parkinson's disease trial. NPJ Parkinsons Dis [Internet]. 2017;3(1). Available from: http://dx.doi.org/10.1038/s41531-017-0013-5

31. Lee DJ, Dallapiazza RF, De Vloo P, Lozano AM. Current surgical treatments for Parkinson's disease and potential therapeutic targets. Neural Regen Res. 2018;13(8):1342–5.

32. Olanow CW, Brin MF, Obeso JA. The role of deep brain stimulation as a surgical treatment for Parkinson's disease. Neurology. 2000;55(12 Suppl 6):S60-6.

33. Wang L, Zhang Q, Li H, Zhang H. SPECT molecular imaging in Parkinson's disease. J Biomed Biotechnol. 2012;2012:412486.

34. Loane C, Politis M. Positron emission tomography neuroimaging in Parkinson's disease. Am J Transl Res. 2011;3(4):323–41.

35. Heim B, Krismer F, De Marzi R, Seppi K. Magnetic resonance imaging for the diagnosis of Parkinson's disease. J Neural Transm (Vienna). 2017;124(8):915–64. .

36. Jones DR, Moussaud S, McLean P. Targeting heat shock proteins to modulate α-synuclein toxicity. Ther Adv Neurol Disord. 2014;7(1):33–51