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Metal Profile and Apolipoprotein E in Parkinson's Disease: Biomarkers of Neurodegenerative Diseases

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ABSTRACT

Background: Iron plays a key role in the pathogenesis of Parkinson's disease (PD), which is the second most frequent neurodegenerative disease. Disturbances in the iron homeostasis can lead to cellular damage through hydroxyl radical production, leading to decreased myelin production and the synthesis and production of neurotransmitters in the central nervous system (CNS).

Aim and Objectives: The aim of the study was to compare the serum levels of iron, copper, ferritin, ceruloplasmin, transferrin, and TIBC levels in PD cases and controls and identify biomarker pool including demographic & genetic variants and blood biochemical markers. Further the association of APOE ε4 allele with PD was also studied.

Materials and Methods: A case-control study was undertaken in 100 PD and non-PD subjects each. Serum iron, copper, ferritin, transferrin, TIBC, ceruloplasmin, and APOE genotyping were measured. Biochemical assessments were performed in Biochemistry autoanalyzers, and APOE genotype was conducted by ARMS-PCR.

Result: In PD subjects serum iron, copper, ferritin, transferrin saturation, and ceruloplasmin levels were significantly low as compared to controls ($p<0.001$) whereas transferrin and TIBC levels were higher as compared to non-diseased subjects ($p<0.01$). The most common APOE genotype observed was ε3ε3 (62%) followed by ε3ε4 (25%) in PD subjects as compared to non-diseased subjects (ε3ε3 89%, ε3ε4 11% respectively). APOE ε4 allele was present in 32.0% PD subjects, as compared to 11% in non-diseased subjects.

Conclusion: In the present study raised iron, copper and APOE ε4 allele are associated with PD, suggesting their potential role as biomarkers in disease risk and progression.

KEYWORDS: Parkinson's disease, Iron profile, Copper, Transferrin, Ferritin, Ceruloplasmin, APOE polymorphism.

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INTRODUCTION

Parkinson's disease (PD) is the second most prevalent neurodegenerative disease of the neuromotor system, affecting 2% world's population. PD patients usually present, with symptoms like bradykinesia (slow movements), rigidity, mask-like facies, slow, rhythmic involuntary tremors, intention tremors etc around at the age of 60 years. Other less frequent presentations are sleep disturbances, gastrointestinal symptoms and psychiatric issues like depression and anxiety affecting other organ systems [1].
Metals play a vital role in the pathogenesis of PD as long term exposure to heavy metals such as lead, mercury, copper, manganese, aluminium, zinc etc. leads to increased risk of PD by impairing the dopamine pathway [2]. These heavy metals cause destruction of the neurons through oxidative stress, mitochondrial dysfunction, protein, endoplasmic reticulum (ER) stress and activation of apoptosis leading PD [3].

Among metals, iron is present in highest concentration in brain as well as whole body and plays an important cofactor in essential functions like DNA synthesis, mitochondrial biogenesis and transportation of oxygen, along with it's role in regulating the synthesis of neurotransmitters, myelin sheath formation, growth and repairing of dendrite spines in hippocampus in brain. In healthy adults, the concentration of iron is heterogeneously distributed in brain being more in basal ganglia (putamen, globus

pallidus, caudate). Changes in the iron level in brain can lead to cognitive decline due to reduced myelination of neurons. The toxicity produced by the iron accumulated in the substantia nigra is due to it's role as cofactor in tyrosine hydroxylase enzyme which limits the neurotransmitters production, therapy thereby contributing to dopamine deficiency. Hence, both excess or shortage of this transition metal can damage dopamine producing neurons along with copper [1].

Copper also plays a dual role, by serving as a coenzyme for crucial antioxidant enzymes like Copper or Zinc SOD (Superoxide Dismutase), leading to the suppression of oxidative stress, whereas as free copper moiety participates in α-synuclein protein oligomerization, oxidative stress and Fenton and Haber-Weiss processes thereby contributing to the formation of Lewy bodies [4].

Transferrin, a transporter of iron produced in liver, attaches two ferric ions forming an apotransferrin Fe3+ complex, transporting them to various tissues through the circulatory system. In addition to being a soluble ferric iron transporter, it also attenuates the redox activity of iron, preventing it from interacting with other molecules. Lower transferrin saturation along with high transferrin and TIBC levels indicate an iron deficiency, thereby increasing the chance of developing Parkinson's disease [5].

The ceruloplasmin, a glycoprotein, transports copper and transforms toxic ferrous (Fe) ions into non-toxic ferric (Fe^{3+}) ions, which it then incorporated into transferrin. Reduced ferroxidase activity of ceruloplasmin results in iron overload in the plasma of individuals with Parkinson's disease [6].

Apolipoprotein E (APOE), a polymorphic protein featuring three primary isoforms (ε2, ε3, and ε4), exists in elevated concentrations within the brain [7]. Various epidemiological studies show APOE as a susceptibility gene for late-onset AD, where APOE ε2 has been implicated zas the protective allele, whereas APOE ε4 has been observed as a risk factor in both AD and PD However these results have been quite conflicting [8]. The APOE gene with three alleles, possessed six genotypes (ε2/ε2, ε3/ε3, ε4/ε4, ε3/ε2, ε4/ε2, and ε4/ε3). Worldwide, APOE ε3 genotype has been found to be most common in almost all the population, followed by the ε4 and ε2 alleles [9]. Literature shows that APOE ε4 has been involved not only in pathogenesis but also in cognitive decline associated with progression of diseases in patients of PD. Such studies are not currently available focusing on the North Indian population. Hence further research in this area is urgently required to understand the potential role of APOE in PD susceptibility and progression within this specific population. The present study was designed to assess the levels of heavy metals like Iron, copper and metalloproteins like TIBC (Total iron binding capacity) ceruloplasmin, Transferrin and Ferritin in Parkinson's disease subjects, along with the frequency of APOE ε4 allele.

METHODOLOGY

Study Design:

A Case Control study was performed in 100 PD and 100 Non-PD subjects in the Department of Neurochemistry, Institute of Human Behaviour and Allied Sciences (IHBAS), New Delhi, India and Department of Biochemistry and Santosh Deemed to be University, Ghaziabad, Uttar Pradesh, India, in collaboration with the Neurology department at IHBAS. Diagnosis of the patients was done by the Neurologist in movement disorders OPD of Neurology department by using the Mini Mental State Examination (MMSE) and Unified Parkinson's Disease Rating Scale (UPDRS). The non-PD subjects with no history of neurovascular surgery, stroke, brain injury, depression or any other mental disorder were included as control subjects. The written consent was obtained from all the PD and non-PD subjects. Iron, copper, iron profile, ceruloplasmin along with APOE genotyping was done in all the subjects.

Biochemical Assessment:

Non fasting blood sample (10-12 ml) was collected during the study by venipuncture in plain evacuation tube (5-6 ml) and EDTA evacuation tube (5-6 ml) from patients in both the study groups taking all aseptic precautions. Plain evacuation tube samples were centrifuged at 1500 rpm for 15 minutes at room temperature within 30 minutes after collection. After centrifugation, the serum was kept at -20°C for analysis of iron, copper, ceruloplasmin, iron profile (ferritin, transferrin, TIBC and UIBC) along with routine laboratory test on Erba XL 1000 an automated biochemistry autoanalyzer from M/s Transasia Bio-Medicals Pvt Ltd. Serum ferritin was measured by chemiluminescent microparticle immunoassay (CMIA) technique on *SI2000, AN IMMUNOASSAY ANALYSER FROM m/S Abbott Pvt. Ltd*. TIBC was calculated mathematically from the estimated serum transferrin (TRF) using formula: TIBC (µmol/L)= 25.0 × TRF g/L. UIBC was calculated by using formula: UIBC (µmol/L) = TIBC (µmol/L) - Serum Iron (µmol/L). EDTA evacuation tube was kept at 2-4°C for DNA isolation. APOE genotyping was done by ARMS- PCR technique.

Statistical Analysis:

All values were presented as mean ± SD. A two-tailed independent test was performed to analyse the continuous variables. Statistical significance was defined at $p<0.05$

RESULT

Socio-Demographic Characteristics

Table 1 shows Socio-Demographic features like age, gender, habitat, smoking and alcohol. All the cases and controls were age and sex matched.

Biochemical Parameters Analysis

The levels of heavy metals iron and copper along with iron profile observed in serum of both PD and non-PD cases are shown in

Table 2. Serum iron and copper levels were significantly low in PD subjects as compared to the non-PD subjects (p< 0.000; p<0.000 respectively), whereas transferrin levels were significantly high in PD cases (p<0.000). Similarly, TIBC levels were significantly higher in PD subjects compared to controls (p<0.003). However, levels of transferrin saturation, ferritin and ceruloplasmin were significantly low in PD subjects (p<0.000; p<0.000; p<0.000 respectively).

APOE Genotyping

APOE genotyping and APOE alleles have been summarized in Table 3 and 4 respectively in cases and control subjects. ε3ε3 (62%) was present in highest percentage of subjects, whereas ε4ε4 genotype was absent in both groups. As shown in table 4, ε3 (77% in PD; 94.5% in non-PD groups) was the most prominent allele in both the groups, followed by ε4 (16% in PD; 5.5% in non-PD groups).

Table 1: Socio-demographic characteristics of PD and Non-PD subjects.

Variables		**Cases**	**Control**	**P-Value**
		(n=100)	**(n=100)**	
Age	Mean (SD)	59.77 (7.40)	59.08 (7.14)	0.78^{NS}
Gender	Male	60 (60.0%)	60 (60.0%)	1.00 ^{NS}
	Female	40 (40.0%)	40 (40.0%)	
Habitat	Rural	27 (27.0%)	42 (42.0%)	0.07 ^{NS}
	Urban	73 (73.0%)	58 (58.0%)	
Alcohol	NO	80 (80.0%)	70 (70.0%)	0.10 ^{NS}
	YES	20 (20.0%)	30 (30.0%)	
Smoking	NO	79 (79.0%)	82 (82.0%)	0.59 ^{NS}
	YES	21 (21.0%)	18 (18.0%)	
Diet	NON-VEG	45 (45.0%)	42 (42.0%)	0.67 ^{NS}
	VEG	55 (55.0%)	58 (58.0%)	

Independent Samples t- test, p<0.05 (Significant), **highly significant, ^{NS}non-significant

TABLE 2: Metal Profile in PD and Non-PD Subjects

Variables		**Groups**		**P value**
		Cases (n=100)	**Control (n=100)**	
Serum Iron	Mean (SD)	12.27 (4.20)	22.99 (5.53)	0.000**
Serum Copper	Mean (SD)	103.49 (17.64)	134.19 (16.78)	0.000**
Serum Transferrin	Mean (SD)	3.59 (0.70)	3.25 (0.45)	0.000**
Transferrin Saturation	Mean (SD)	14.17 (5.13)	28.76 (8.10)	0.000**
TIBC	Mean (SD)	88.28 (20.18)	81.37 (11.20)	0.003**
Serum Ferritin	Mean (SD)	53.91 (34.10)	99.65 (82.85)	0.000**
Serum Ceruloplasmin	Mean (SD)	31.21 (7.00)	39.50 (7.34)	0.000**

Independent Samples t- test, p<0.05 (Significant), **highly significant, ^{NS} non-significant

Table 3: APOE Genotyping in PD & non-PD cases

APOE Genotype	**CASES**	**CONTROL**
	Frequency (%)	**Frequency (%)**
ε2ε2	1 (1.0)	0 (0.0)
ε2ε3	5 (5.0)	0 (0.0)
ε2ε4	7 (7.0)	0 (0.0)
ε3ε3	62 (62.0)	89 (89.0)
ε3ε4	25 (25.0)	11 (11.0)
ε4ε4	0 (0.0)	0 (0.0)
Total	100 (100.0)	100 (100.0)

Table4: Distribution of APOE Allele in PD & non-PD cases

APOE Allele	**Cases**	**Control**
	Frequency n (%)	**Frequency n (%)**
ε2	14 (7.0)	0 (0.0)

ε3	154 (77.0)	189 (94.5)
ε4	32 (16.0)	11 (5.5)

DISCUSSION

In the present study, significant alterations in the metal profile (low iron, low copper, low ferritin, low ceruloplasmin and elevated transferrin in PD subjects) and differential Apolipoprotein E (APOE) isoform distributions in subjects with PD in contrast to healthy participants were observed. Our findings suggest that both dysregulated metal homeostasis and APOE ε4 allele may play a major role in pathogenesis of PD and can serve as potential biomarkers for neurodegenerative processes in PD.

Iron & Copper in Parkinson's disease

Our results are consistent with previous studies reporting increased accumulation of iron in brain among PD cases, particularly in the substantia nigra, which may contribute to oxidative stress and dopaminergic neuron degeneration. Significant reduction in serum iron due to its buildup in the substantia nigra results in toxicity in PD, as iron acts as a cofactor for tyrosine hydroxylase enzyme, limiting neurotransmitter production leading to dopamine shortage. Thus, in PD dopaminergic neurons may suffer from an excess or deficiency of the transition metal elements, particularly iron and copper [10].

Our findings are supported by other studies done by Gangania et al. [11] reporting decreased serum iron levels in PD subjects as compared to healthy controls Contrarily few studies (12, 13, 14) reported no significant difference of serum iron levels between PD and controls. According to a study by Hedge et al. [15], serum Fe levels dropped as the severity of PD progressed. Furthermore, Pichler et al. [5] reported that increased blood iron levels prevent Parkinson's disease (PD), with a 3% decrease in PD risk for every 10µg/dL increase in serum iron levels.

Evidence shows that iron accumulates in different areas of the brain, especially in the substantia nigra. Consequently, a reduced iron level in the bloodstream is viewed as a risk factor for the onset of PD. As iron acts as a cofactor for tyrosine hydroxylase, which plays a key role in the synthesis of neurotransmitter dopamine, its' reduced levels in substantia nigra result in the dysfunction of neurons.

The pathophysiology of PD is significantly influenced by other heavy metals like copper as well. As a cofactor for the antioxidant enzyme copper-zinc superoxide dismutase, it contributes to both the formation of oxidative stress and its removal. In the present study, the concentration of low copper levels in blood of PD subjects was observed as compared to non-PD group. Similar findings have been reported in the literature as well by many research groups (11, 12, 16, 17). However, no correlation of copper levels has been reported with age, duration of disease, or levodopa consumption. In contrast to this, Hedge et al. [15] have reported elevated serum copper levels in both early and advanced PD cases in their study, indicating that the onset of PD may be associated with reduced serum copper concentrations by its impact on the non-motor symptoms related to PD. However, a meta-analysis performed by Mariana et al. [18] showed no prominent difference in blood and CSF copper levels between PD patients and healthy controls [12]. These findings indicate that there may be a disruption in copper balance in the advanced phase of the illness in PD and the potential involvement of this trace element in Parkinson's disease is suggested by decrease in serum level of copper. Similar to iron, there has been conflicting findings in literature regarding involvement of copper in Parkinson's disease. Forte et al. [19] and Bocca et al. [20] reported lower serum copper levels in PD, whereas Jiménez-Jiménez et al. [21] has reported no significant alteration in serum copper levels in PD as compared to controls. Our findings are also consistent with the histology investigations revealing a 34% reduction in copper in substantia nigra when compared to controls in similar age group [4]. But authors did not note any disruption of copper in the substantia nigra in patients with multisystemic atrophy or supranuclear palsy, indicating that this drop was selective. Additionally, Pall et al. [22] and colleagues reported a rise in the copper in PD patients' cerebrospinal fluid (CSF), a finding which was not supported in subsequent research [17].

Iron Profile in Parkinson's disease

In present study, significantly raised transferrin levels were observed, whereas serum ferritin, ceruloplasmin, TIBC levels and Transferrin saturation were low in PD subjects. In a previous study by Gangania et al. [11] PD patients exhibited increased transferrin and TIBC levels, along with decreased ferritin and transferrin saturation levels and was supported by other studies performed by Farhoudi et al. [23] and Annamaki et al. [24]. Nevertheless, Logroscino et al. [13] and Shen at al. [14] found lower levels of ferritin, transferrin, transferrin saturation and TIBC in PD subjects as compared to non-PD cases, whereas Chen at al. [25] reported higher transferrin and ferritin levels in PD subjects. However, they were not able to find any correlation with the duration of treatment or iron intake from food. Ferritin is present in the cytoplasm of almost all cells. It is a two-subunit, 24-polymer protein whose main biological function is iron storage. So far, as per available research no definite connection between serum ferritin and Parkinson's disease has been established [26]. Similarly Mariani et al. [27] reported high levels of serum transferrin and transferrin saturation in PD subjects, but Xu et al. [28] found that neither transferrin nor its receptor increases in the PD brain. Hence iron buildup in Parkinson's disease may not be due to transferrin or its receptor [26], rather iron triggers a redox mechanism in metabolism that results in the production of hydroxyl free radicals when the level of iron surpasses the transport ability of transferrin [29]. The released free radicals may result in pathological alterations and may play a crucial role in pathogenesis of neurodegenerative diseases, by lipid peroxidation, mitochondrial dysfunction, cellular injury, chronic inflammation, leading to synuclein accumulation in brain [30,31]. In their study Ayton et al concluded that the iron deposition may result from the restriction of iron flux in substantia nigra caused by the loss of transferrin in PD with concomitant rise of iron be [32].

Apolipoprotein E in Parkinson's disease

APOE gene, located on chromosome 19q13.2, codes for protein which facilitates cholesterol transport and thereby aids in brain injury recovery. In research undertaken on AD till date, APOE has been consistently reported to be a probable candidate gene that might influence the neurodegenerative process during the progression of disease. It has also been proposed that there may be numerous pathophysiological similarities between AD and PD as protein aggregation and neuronal death have been observed in both illnesses, leading to cognitive decline and extrapyramidal symptoms as their pathogenomic clinical characteristics. Hence like AD, APOE is considered as a probable candidate gene that might influence the neurodegenerative processes in PD leading to cognitive decline which has been given a separate identity- Parkinson's diseases dementia (PDD). The APOE gene has three alleles: ε2, ε3 and ε4 due to variation in arginine and cysteine presence at amino acid position 112 and 158. Alleles ε3 and ε2 are more efficient in mediating neuronal remodelling, repair, and protection than the ε4 allele [33]. As ε4 has arginine at 112, by virtue of which it produces a property referred to as Domain interaction (ε4 > ε3 > ε2). Hence, APOE ε4 isoform is identified as being substantially linked to the risk of AD, LOAD, PD, and PDD out of the three alleles (ε2, ε3, and ε4). APOE ε3 is neutral, whereas APOE ε2 is usually considered to be protective against neurodegenerative illnesses [9].

As shown in Table 3, in the present study the APOE ε4 allele was observed in 32 PD subjects (16%) whereas only 11 non-PD subjects (5.5%) had ε4. Tripathi et al. [34] reported 25% PD subjects with atleast one ε4 allele as compared to control (11.5%). Furthermore, same group also reported APOE ε4 as a significant risk factor in AD and PD groups, contributing more than 2.5 times risk of having disease as compared to non-diseased group, Similarly, in another study performed by Ghebremedhin et al. [35] on moderate to severe PD had increased frequency of APOE ε4 allele with decreased APOE ε3 allele. In contrast, meta-analysis performed with 22 association studies of PD showed APOE ε2 as a risk factor for PD with no effect of APOE ε4 [36]. Hence it can be concluded that APOE ε4 is a significant genetic influence in the progression of PD, but its role needs to be further investigated to collect more evidences.

CONCLUSION

Iron dysregulation (especially ferritin and TIBC changes) is consistently linked to Parkinson's disease pathophysiology but iron in CSF can also be seen for better results. Ceruloplasmin deficiency may aggravate the iron accumulation in the brain due to its oxidase activity. In addition to it, copper imbalance seems to play a role in pathophysiology through oxidative damage. These markers are promising for early diagnosis of PD and therapeutic targets.

REFERENCES

1. Raj K, Kaur P, Gupta G.D. Singh S. Metals associated neurodegeneration in Parkinson's disease: Insight to physiological, pathological mechanisms and management. Neuroscience Letters. 2021; 753:135873.
2. Tchounwou PB, Yedjou CG, Patlolla AK, Sutton DJ, Heavy metal toxicity and the environment. Mol Clin and Environ Toxic. 2012;133-164.
3. Ball N, Teo WP, Chandra S, Chapman J. Parkinson's Disease and the environment. Front. Neurol. 2019; 10:31-8.
4. Gangania MK, Batra J, Kushwaha S, Agarwal R. Role of Iron and Copper in the Pathogenesis of Parkinson's Disease. Ind J Clin Biochem. 2016; DOI 10.1007/s12291-016-0614-5
5. Pichler I, Fabiola Del Greco M, Gögele M, Lill CM, Bertram L, Do CB, Eriksson N, Foroud T, Myers RH, Nalls M, Keller MF. Serum iron levels and the risk of Parkinson disease: a mendelian randomization study. PLoS Med. 2013 Jun 4; 10(6): e1001462.
6. Jin L, Wang J, Zhao L, Jin H, Fei G, Zhang Y, et al. Decreased serum ceruloplasmin levels characteristically aggravate nigral iron deposition in Parkinson's disease. Brain. 2011;134(1):50–8.
7. Mahley R. Apolipoprotein E: Cholesterol transport protein with expanding role in cell biology. Science. 1988; 240:622–30.
8. Gaoa J, Huangb X, Parkc Y, Liua R, Hollenbeckd A, Schatzkin A et al. Apolipoprotein E genotypes and the risk of Parkinson disease. Neurobiol Aging. Nov 2011; 32(11): 2106.e1–2106.e6.
9. Li J, Luo J, Liu L, Fu H, Tang L. The genetic association between apolipoprotein E gene polymorphism and Parkinson disease: A meta-analysis of 47 studies. Medicine. 2018; 97:43.
10. Oakley AE, Collingwood JF, Dobson J, Love G, Perrott HR, Edwardson JA, et al. Individual dopaminergic neurons show raised iron levels in Parkinson disease. Neurology. 2007;68(21):1820–5.
11. Gangania MK, Batra J, Kushwaha S, Agarwal R. Impaired Systemic Iron Homeostasis and Parkinson's Disease. JMSCR. 2017;05(04):20855-9.
12. Asad T, Aamir M, Haroon ZH, Munir MU, Kirmani SI, Awan A. Comparison of Serum Copper, Zinc, Lead, Aluminium and Iron Levels in Patients with Parkinson's Disease with Healthy Controls in Tertiary Care Hospital. Pak Armed Forces Med J 2022;72(5):1673.
13. Logroscino G, Marder K, Graziano J, Freyer G, Slavkovich V, Lolacono N, et al. Altered systemic iron metabolism in Parkinson's disease. Neurology. 1997;49(3):714–7.
14. Shen X, Yang H, Zhang D, Jiang H. Iron Concentration Does Not Differ in Blood but Tends to Decrease in Cerebrospinal Fluid in Parkinson's Disease. Frontiers in Neuroscience. Sep 2019; 13(939):1-11.
15. Hegde ML, Shanmugavelu P, Vengamma B, Rao TS, Menon RB, Rao RV, et al. Serum trace element levels and the complexity of inter-element relations in patients with Parkinson's Neurology. disease. J Trace Elem Med Biol. 2004; 18(2):16371.
16. Ilyechova EY, Miliukhina IV, Orlov IA, Muruzheva ZM, Puch kova LV, Karpenko MN. A low blood copper concentration is a co-morbidity burden factor in Parkinson's disease development. J Pers Med. 2019; 9(3):41.

17. Samia YM, Mouna A, Narjes M, Amel BB, Saber C, Mahbouba FA et al. serum Copper, Zinc and selenium levels in Tunisian patients with Parkinson's disease. 2013; 91:402-5.
18. Mariani S, Ventriglia M, Simonelli I, Donno S, Bucossi S, Vernieri F, et al. Iron and Copper do not differ in Parkinson's disease: a replication study plus meta-analysis. Neurobiol Aging. 2013; 34(2):632-23.
19. Forte G, Bocca B, Senofonte O, et al. Trace and major elements in whole blood, serum, cerebrospinal fluid and urine of patients with Parkinson's disease. J Neural Transm 2004; 111:1031- 40.
20. Bocca B, Alimonti A, Senofonte O, et al. Metal changes in CSF and peripheral compartments of parkinsonian patients. J Neurol Sci.2006; 248:23–30.
21. Jimenez-Jimenez FJ, Molina JA, Aguilar MV, Meseguer I, Mateos-Vega CJ, Gonzalez-Munoz MJ, et al. Cerebrospinal fluid levels of transition metals in patients with Parkinson's disease. J Neural Transm. 1998; 105(45):497–505.
22. Pall HS, Williams AC, Blake DR, et al. Raised cerebrospinal-fluid cooper concentration. Lancet 1987;2(8553):238-41.
23. Farhoudi, M., Taheraghdam, A., Farid, G. A., Talebi, M., Pashapou, A., Majidi, J., et al. Serum iron and ferritin level in idiopathic Parkinson. Pak. J. Biol. Sci. 2012; 15:1094–7.
24. Annanmaki T, Muuronen A, Murros K. Low plasma uric acid level in Parkinson's disease. Mov Disord.2007;22:1133-7.
25. Chen Zt, Pan Cz, Ruan Xl, Lei Lp, Lin Sm, Wang Yz, et al. Evaluation of ferritin and TfR level in plasma neural-derived exosomes as potential markers of Parkinson's disease. Front. Aging Neurosci. Sep 2023; 15:1216905.
26. Tripathi CB, Gangania MK, Kushwaha S, Agarwal R. Evidence-Based Discriminant Analysis: A New Insight into Iron Profile for the Diagnosis of Parkinson's Disease.Annals of Indian Academy Neurology. 2021; 24(2);234-8.
27. Mariani S, Ventriglia M, Simonelli I, Donno S, Bucossi S, Vernieri F, et al. Fe and Cu do not differ in Parkinson's disease: A replication study plus meta-analysis. Neurobiol Aging. 2013; 34:632-3.
28. Xu HM, Jiang H, Wang J, Luo B, Xie JX. Over-expressed human divalent metal transporter 1 is involved in iron accumulation in MES23.5 cells. Neurochem Int.2008; 52:1044-51.
29. Youdim MB, and Riederer P. The role of iron in senescence of dopaminergic neurons in Parkinson's disease. J. Neural Transm. Suppl.1993; 40:57–67.
30. Gutteridge JM. Hydroxyl radicals, iron, oxidative stress, and neurodegeneration. Ann. N. Y. Acad. Sci. 1994; 738:201–13.
31. Sian-Hulsmann J, MandelS, Youdim, MB, and Riederer, P. The relevance of iron in the pathogenesis of Parkinson's disease. J. Neurochem.2011; 118:939–57.
32. Ayton S, Lei P, Mclean C, Bush A, Finkelstein D. Transferrin protects against Parkinsonian neurotoxicity and is deficient in Parkinson's substantia nigra. Signal Transduction and Targeted Therapy. 2016; 1:16015.
33. Singh NK, Banerjee BD, Bala K, Mitrabasu A, Dung AA, Chhillar N. APOE and LRPAP1 gene polymorphism and risk of Parkinson's disease. Neurol Sci. 2014; 35:1075–81.
34. Moreau C, Duce JA, Rascol O, Devedjian JC, Berg D, Dexter D et al. Iron as a Therapeutic Target for Parkinson's Disease. Movement Disorders. 2018; 33(4):568-74.
35. Ghebremedhin E, Del Tredici K, Vuksic M, Rüb U, Thal DR, et al. Relationship of apolipoprotein E and age at onset to Parkinson disease neuropathology. J Neuropathol Exp Neurol. 2006; 65:116-123.
36. Huang X, Chen PC, Poole C. APOE-epsilon2 allele associated with higher prevalence of sporadic Parkinson disease. Neurology. 2004; 62:2198-2202.

Declarations

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Ethical approval

The study protocol was reviewed and approved by the Institutional Ethics Committee of Institute of Human Behaviour and Allied Sciences (IHBAS), New Delhi, India, Approval No.: IEC-3/IHBAS/January 2024/ 4. All procedures involving human participants were performed in accordance with the ethical standards of the institutional and/or national research committee and with the 1964 Helsinki Declaration and its later amendments. Written informed consent was obtained from all participants prior to sample collection.

Consent to participate

All the subjects voluntarily agree to participate in this research. Written informed consents were provided by all the subjects participated in the study.

Consent for publication

All authors reviewed the results and approved the final version of the manuscript.

Authors' contributions

Author 1 has written the paper and performed the research i.e., collected and processed the samples as it is her thesis topic of PhD. Author 2 has designed the study, data analysis and helped in the interpretation and manuscript drafting. Author 3 has diagnosed the patients from the OPD. Author 4 has analysed the data and done statistics. Author 5 has helped in analysis of data, interpretation and manuscript drafting. Author 1, 2 & 5 has critically revised the manuscript for intellectual content. All authors

read and approved the final version of the manuscript and agreed to be accountable for all aspects of the work.

Data availability

All relevant data can be found within the manuscript.

Competing interests

The authors declare that they have no competing interests.

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List of Abbreviations:

AD: Alzheimer's Disease
APOE: Apolipoprotein E
ARMS-PCR: Amplification Refractory Mutation System- Polymerase Chain Reaction
CMIA: Chemiluminescent Microparticle Immunoassay
CSF: Cerebrospinal Fluid.
DNA: Deoxyribonucleic Acid
EDTA: Ethylenediaminetetraacetic acid
ER: Endoplasmic Reticulum
Fe2+: ferrous ion
Fe3+: ferric ion
LOAD: Late-Onset Alzheimer's Disease
MMSE: Mini-Mental State Examination
OPD: Outpatient Department
PDD: Parkinson's Diseases Dementia
Rpm: Revolutions Per Minute
SOD: Superoxide Dismutase
TIBC: Total Iron-Binding Capacity
TRF: Transferrin
UIBC: Unsaturated Iron Binding Capacity
UPDRS: Unified Parkinson's Disease Rating Scale