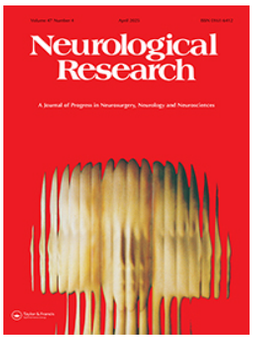

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Serum neurofilament light chain as a potential biomarker in restless legs syndrome: a cross-sectional study

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ABSTRACT

Objective: Neurofilament light chain (NfL) has emerged as a promising biomarker for several neurological diseases. Therefore, we investigated its serum levels and their association with disease characteristics, including duration, severity, and medication use in patients with restless legs syndrome (RLS).

Method: This cross-sectional prospective study included 71 RLS patients and 70 healthy controls. RLS patients were characterized based on disease duration, severity, and medication use. NfL levels were quantified using commercial enzyme-linked immunosorbent assay kits.

Result: No significant differences in NfL levels were observed between RLS patients and controls ($p > 0.05$). Furthermore, the levels were not significantly associated with disease duration or severity in RLS patients ($p > 0.05$).

Conclusion: These findings do not support the use of NfL as a biomarker for RLS. Further large-scale studies are needed to evaluate its role in RLS.

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Neurofilament light chain; restless legs syndrome; neurodegeneration; Willis–Ekbom disease; biomarker

Introduction

Restless legs syndrome (RLS), also known as Willis – Ekbom disease, is a common sensory-motor disorder characterized by unpleasant sensations in the legs and an irresistible urge to move the limbs [1]. With a prevalence of 5–10% in adults from North America and Western Europe, RLS is one of the most common sleep disorders [2]. Notably, the condition is twice as common in women, and its incidence increases with age [3]. The primary form, known as idiopathic RLS, is the most common type that presents early in life, peaking during the second decade; it has an underlying genetic predisposition and accounts for approximately 70–80% of all RLS cases [3]. By contrast, secondary RLS develops later in life and is associated with underlying medical conditions, including pregnancy, chronic kidney disease, rheumatic diseases, certain medications, diabetes mellitus, Parkinson's disease (PD), and multiple sclerosis (MS) [2,3]. Despite extensive research, the underlying pathophysiology of primary RLS remains uncertain. However, genetic predisposition, central nervous system iron dysregulation, and dopaminergic dysfunction are implicated as potential etiological factors [2].

Neurofilament light chain (NfL) has emerged as a promising biomarker for several neurological diseases [4]. Elevated levels have been reported in patients with PD [5,6], MS [7–9], Alzheimer's disease (AD) [10,11], motor neuron disease [12,13], and traumatic brain injury [14,15]. In particular, NfL is poised to become a critical diagnostic criterion for MS [16,17] and AD [18,19] in future guidelines. As a core component of the neuronal cytoskeleton, NfL is released in small quantities into the blood and cerebrospinal fluid (CSF), with levels increasing with age [20]. However, axonal damage or neuronal degeneration leads to significantly increased NfL release into the interstitial fluid of the brain, entering the blood and CSF [21]. Although traditionally measured in CSF, recent advancements in ultrasensitive assays have enabled the detection of NfL in blood, a less invasive sample [22]. Vrillon et al. [23] demonstrated the equivalent diagnostic performance of NfL levels measured in the CSF and plasma for the positive and differential diagnosis AD in a clinical setting.

Here, we investigated serum levels of NfL and their association with disease characteristics, including duration, severity, and medication use in patients with RLS.

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Method

Study population

A cross-sectional prospective study was conducted between March and September 2023 at Kirsehir Ahi Evran University Training and Research Hospital in Kirsehir, Turkey. We included 71 patients with idiopathic RLS and 70 age- and sex-matched healthy controls aged >18 years. Patients with malignancies, neurodegenerative diseases (AD, PD, and MS), a history of local trauma or surgery, peripheral nervous system pathologies including neuropathies or radiculopathies, and pregnancy were excluded.

Idiopathic RLS was diagnosed in accordance with the International RLS Study Group diagnostic criteria [24]. RLS patients underwent evaluation of disease duration, severity, and medication use. Disease duration was categorized as newly – diagnosed, <5, 5–9, and ≥10 years. Disease severity was evaluated using the International RLS Study Group severity scale, ranging from 0 to 40 points. Scores of 0–10, 11–20, 21–30, and 31–40 represent mild, moderate, severe, and very severe RLS, respectively [25]. Dopamine agonists were prescribed to 84.5% of RLS patients. Fasting venous blood samples were collected from all patients, and standard laboratory analyses, including complete blood count, iron studies (iron, total iron binding capacity, ferritin, and transferrin saturation), vitamin B12, folate, vitamin D, and magnesium, were conducted.

The Kirsehir Ahi Evran University Local Research Ethics Committee (approval date 1 March 2023; approval no. 2023–01/01) approved the study protocol. Informed consent was obtained from all participants.

NfL assay

Blood samples were collected from RLS patients and controls in gel tubes without anticoagulants and centrifuged at 3000 rpm for 10 min. Subsequently, the supernatant was removed and stored at −80°C until analysis. Serum concentrations of NfL were determined using a commercially available enzyme-linked immunosorbent assay (ELISA) kit (Elabscience, Wuhan, China). Optical density was measured at 450 nm using a microplate reader (SPECTROstar Nano, BMG Labtech, Ortenberg, Germany); NfL concentrations were expressed as pg/mL.

Statistical analysis

Statistical analyses were performed using SPSS software (ver. 25.0; IBM Corp., Armonk, NY, U.S.A.). Normality was assessed using the Kolmogorov – Smirnov test. Data are presented as mean ± standard deviation, median (interquartile range), or percentages. Continuous variables were compared using a one-way analysis of variance, Kruskal – Wallis test, Student's t-test, or Mann – Whitney U test, whereas categorical variables were compared using the chi-square test. Pearson correlations were calculated to assess relationships between numerical variables. p-values < 0.05 were considered statistically significant.

Result

Table 1 presents the baseline demographics and laboratory characteristics of the control group and RLS patients. The median age of RLS patients was 56 (23–69) years, with female predominance (80.3%). No significant differences in age, sex, laboratory parameters, or NfL levels were observed between the groups ($p > 0.05$).

Table 2 presents the blood parameters of RLS patients categorized by disease duration. Of the 61 patients, 11 (15.5%) were newly diagnosed, 28 (39.4%) had RLS for <5 years, 14 (19.7%) for 5–9 years, and 18 (25.3%) for ≥10 years. No significant differences in laboratory parameters or NfL levels were observed among the groups ($p > 0.05$).

Table 3 presents the blood parameters of RLS patients categorized by disease severity. Based on the International RLS scale, no patients exhibited mild RLS, whereas 26 (36.6%) had moderate, 42 (59.2%) had severe, and 3 (4.2%) had very severe RLS. Considering the heterogeneity of the subgroups, patients were dichotomized into severe ($n = 45$, 63.4%) and non-severe ($n = 26$, 36.6%) groups. No significant differences in blood parameters or NfL levels were observed between these groups ($p > 0.05$).

Table 4 presents the correlation analysis of blood parameters in RLS patients. Of these parameters, NfL exhibited a significant correlation with magnesium ($r = 0.210$, $p = 0.012$).

Discussion

The pathophysiology of RLS remains uncertain, hindering diagnosis, treatment, and prognosis. Proposed mechanisms include genetic predisposition, dopaminergic dysfunction, iron metabolism disturbances, and central opioidergic system dysfunction [2]. Considering the high prevalence of familial RLS (>60%), genetic factors may primarily contribute to RLS pathogenesis. Multiple studies have elucidated the

Table 1. The demographic and laboratory data of controls and patients with restless legs syndrome.

	Controls ($n = 70$)	Patients ($n = 71$)	P–value
Age (years)	53 (20 – 71)	56 (23 – 69)	0.256
Gender			
Female	47 (67.1)	57 (80.3)	0.087
Male	23 (32.9)	14 (19.7)	
Blood parameters			
Iron (µg/dL)	78.3 ± 39.2	68 ± 35.7	0.126
TIBC (µg/dL)	363.9 ± 54.3	375.2 ± 68	0.278
Ferritin (ng/mL)	45.5 (6 – 495)	39 4 – 793)	0.689
Transferrin saturation (%)	22.4 ± 13.0	18.7 ± 10.3	0.148
Haemoglobin (g/dL)	14 ± 1.7	13.9 ± 1.7	0.746
Vitamin B12 (pg/mL)	381.6 ± 122.6	357.5 ± 115.9	0.295
Folate (ng/mL)	7 (2 – 16)	7 (3 – 20)	0.852
Vitamin D (ng/mL)	17 (5 – 51)	16 (5 – 78)	0.248
Magnesium (mg/dL)	2 (1.4 – 2.3)	2 (0.9 – 2.4)	0.536
NfL (pg/mL)	12.5 (7.6 – 97.8)	12.3 (7.7 – 39.5)	0.779

Values are expressed as n (%), mean ± SD or median (min – max). TIBC indicates total iron – binding capacity; NfL, neurofilament light chain.

Table 2. Blood parameters of patients with restless legs syndrome according to disease duration.

	Newly – diagnosed ($n = 11$)	<5 years ($n = 28$)	5–9 years ($n = 14$)	≥10 years ($n = 18$)	P–value
Iron (µg/dL)	85 ± 55.1	68.1 ± 31.9	65.5 ± 32.4	59.4 ± 27.8	0.699
TIBC (µg/dL)	367.3 ± 70.3	387.6 ± 68.8	376.4 ± 53.9	359.7 ± 76.5	0.448
Ferritin (ng/mL)	65 (6 – 200)	38 (4 – 255)	35 (6 – 793)	34 (5 – 112)	0.583
Transferrin saturation (%)	24.6 ± 17.0	18.5 ± 9.4	17.4 ± 7.7	16.6 ± 6.9	0.629
Haemoglobin (g/dL)	13.7 ± 2.5	13.9 ± 1.4	14.0 ± 1.9	13.9 ± 1.6	0.997
Vitamin B12 (pg/mL)	338 ± 123.3	364.4 ± 105.5	332.3 ± 80.1	378.2 ± 149.9	0.703
Folate (ng/mL)	6 (3 – 13)	8 (4 – 20)	5.5 (3 – 20)	8 (3 – 20)	0.088
Vitamin D (ng/mL)	15 (8–28)	15 (6 – 51)	16 (8 – 28)	18 (5 – 78)	0.536
Magnesium (mg/dL)	2 (1.5 – 2.3)	2 (1.9 – 2.3)	2 (1.9 – 2.2)	2 (0.9 – 2.4)	0.810
NfL (pg/dL)	13.5 (7.8–30.7)	11.2 (7.7 – 33.5)	13.7 (7.9 – 39.5)	13.2 (7.9 – 34.3)	0.512

Values are expressed as mean ± SD or median (min – max). TIBC indicates total iron – binding capacity; NfL, neurofilament light chain.

Table 3. Blood parameters of patients with restless legs syndrome in terms of disease severity.

	Severe ($n = 45$)	Non – severe (n=26)	P–value
Iron (µg/dL)	67 ± 33.7	69.6 ± 39.7	0.213
TIBC (µg/dL)	377.2 ± 72.3	371.6 ± 61.1	0.298
Ferritin (ng/dL)	50[4–793]	37.5[5–270]	0.914
Transferrin saturation (%)	18.5 ± 9.7	19.1 ± 11.4	0.404
Haemoglobin (g/dL)	13.9 ± 1,8	13.9 ± 1.6	0.539
Vitamin B12 (pg/mL)	361.2 ± 113.5	351.0 ± 122.0	0.265
Folate (ng/mL)	7 [3 – 20]	7[3 – 13]	0.710
Vitamin D (ng/mL)	15[6 – 78]	16 [5 – 51]	0.938
Magnesium (mg/dL)	2 [0.9 – 2.3]	2 [1.5 – 2.4]	0.646
NfL (pg/dL)	12.3 [7.7 – 39.5]	12.7[7.7 – 34.3]	0.877

Values are expressed as mean ± SD or median [min – max]. TIBC indicates total iron – binding capacity; NfL, neurofilament light chain.

underlying genetic mechanisms of RLS. Genome-wide association studies in Northern European populations have identified several susceptibility genes, including BTBD9, MEIS1, PTPRD, MAP2K5/SKOR1, and TOX3/BC034767 [26–28]. Schormair et al. [29] identified 19 risk genes associated with RLS pathogenesis. Although low iron levels can contribute to RLS symptoms, most patients, including those in our study, exhibit normal iron tests. Several studies have indicated that iron deficiencies in the brain, rather than systemic depletion, is a critical factor in RLS pathogenesis [30]. Transcranial ultrasound examinations by Schmidauer et al. [31] demonstrated decreased iron content in the substantia nigra, a primary iron reservoir in the brain, among RLS patients. Subsequent magnetic resonance imaging (MRI) studies have corroborated these findings [32]. Iron deficiencies in the brain are implicated in the

Table 4. Correlation analysis of blood parameters in patients with restless legs syndrome.

	Iron	TIBC	Ferritin	Transferrin saturation	Hemoglobin	Magnesium	NfL
Iron	.	r= −0.213 $p = 0.011$	r = 0.544 $p < 0.001$	r = 0.926 $p < 0.001$	r = 0.530 $p < 0.001$	NS	NS
TIBC	r = −0.213 $p = 0.011$	.	r = 0.536 $p < 0.001$	r = −0.419 $p < 0.001$	r = −0.312 $p < 0.001$	NS	NS
Ferritin	r = 0.544 $p < 0.001$	r = 0.536 $p < 0.001$	.	r = 0.625 $p < 0.001$	r = 0.567 $p < 0.001$	NS	NS
Transferrin saturation	r = 0.926 $p < 0.001$	r = −0.419 $p < 0.001$	r = 0.625 $p < 0.001$	.	r = 0.526 $p < 0.001$	NS	NS
Hemoglobin	r = 0.530 $p < 0.001$	r = −0.312 $p < 0.001$	r = 0.567 $p < 0.001$	r = 0.526 $p < 0.001$	.	r = 0.235 $p = 0.005$	NS
Magnesium	NS	NS	NS	NS	r = 0.235 $p = 0.005$	.	r = 0.210 $p = 0.012$
NfL	NS	NS	NS	NS	NS	r = 0.210 $p = 0.012$	.

TIBC indicates total iron – binding capacity; NfL, neurofilament light chain.

pathophysiology of RLS, contributing to hypoxia and myelin loss [33]. Previous imaging studies have demonstrated increased putaminal dopaminergic stimulation in response to hypoxia in RLS patients [34]. Consequently, synaptic dopamine levels rise, leading to postsynaptic downregulation of receptor and cellular function in the brain [33]. Considering the circadian fluctuations in dopamine, this postsynaptic adaptation remains asymptomatic during the day but becomes apparent at night when dopamine levels decrease, resulting in RLS symptoms. This mechanism underlies the efficacy of levodopa for treating nocturnal RLS symptoms [30].

NfL is a cytoplasmic protein predominantly expressed in large-caliber myelinated axons. Serum levels correlate with axonal damage severity across various neurological disorders, including neurodegenerative, inflammatory, and traumatic disorders [6,7,10,13,15]. Recent studies have predominantly focused on PD [5,6,35], MS [7,8,17], and AD [10,11,36]. Diekamper et al. [37] demonstrated a strong correlation between serum levels NfL and reduced presynaptic dopamine transporter density in the putamen of patients with movement disorders and nigrostriatal degeneration. These findings suggest that NfL may serve as a biomarker for motor function impairment in PD. In addition, Teunissen et al. [38] reported elevated CSF and serum levels in patients with MS and clinically isolated syndrome who subsequently developed MS. Mattsson et al. [36] conducted a longitudinal analysis of plasma levels in 1,583 North American AD patients, collecting data annually for 11 years (2005–2016). Their findings established NfL in plasma as a noninvasive biomarker for monitoring neurodegeneration in AD. Moreover, several studies have investigated serum levels of NfL in COVID-19 patients, a condition characterized by excessive inflammation. Although several studies have reported elevated levels in such patients with neurological symptoms [39,40], others have found no significant differences [41]. In addition, several studies have implicated NfL in the neuronal death associated with neurodegeneration in RLS patients [42,43]. Consequently, we hypothesized that NfL levels would be elevated in RLS patients. However, no significant differences were observed between RLS patients and controls, irrespective of disease severity or duration. Several factors may explain these unexpected results. First, the small sample size may have limited our ability to detect significant differences. Second, the inclusion of RLS patients with idiopathic disease, characterized by substantial genetic heterogeneity in neuronal protein expression, could have introduced variability in NfL levels. In addition, racial differences may have confounded our findings. Moreover, most patients (84.5%) were treated with dopamine agonists, which decreased NfL levels and attenuated potential differences between groups. Although serum levels of NfL did not show significant differences between groups, CSF analysis may provide additional insights.

Our study had several limitations. Notably, the small sample size, cross-sectional design, and the prevalence of medication use among participants may have influenced our findings. Additionally, we measured serum levels of NfL using ELISA whereas a more sensitive assay such as single-molecule array (Simoa) might increase the sensitivity in such a neurological disorder without a major neurodegenerative component. However, a recent comparative study has shown similar efficacy for both, ELISA and Simoa, in analyzing plasma levels of NfL [44].

Conclusion

Our study is the first to evaluate serum levels of NfL as a biomarker in RLS patients. Although our findings do

not currently support the use of NfL as a biomarker for RLS, larger-scale studies are needed to evaluate its role in RLS.

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References

[1] Khachatryan SG, Ferri R, Fulda S, et al. Restless legs syndrome: over 50 years of European contribution. J Sleep Res. 2022;31(4):e13632. doi: 10.1111/jsr.13632
[2] Khan FH, Ahlberg CD, Chow CA, et al. Iron, dopamine, genetics, and hormones in the pathophysiology of restless legs syndrome. J Neurol. 2017;264(8):1634–1641. doi: 10.1007/s00415-017-8431-1
[3] Ghorayeb I, Tison F. Épidémiologie du syndrome des jambes sans repos [Epidemiology of restless legs syndrome]. Rev Neurol (Paris). 2009;165(8–9):641–649. doi: 10.1016/j.neurol.2009.02.006
[4] Barro C, Zetterberg H. Neurological symptoms and blood neurofilament light levels. Acta Neurol Scand. 2021;144(1):13–20. doi: 10.1111/ane.13415
[5] Bäckström D, Linder J, Jakobsonet al. NfL as a biomarker for neurodegeneration and survival in Parkinson disease. Neurology. 2020;95(7):e827–e38. doi: 10.1212/WNL.0000000000010084
[6] Buhmann C, Magnus T, Choe CU. Blood neurofilament light chain in Parkinson's disease. J Neural Transm (Vienna). 2023;130(6):755–762. doi: 10.1007/s00702-023-02632-7
[7] Abdelhak A, Benkert P, Schaedelin S, et al. Neurofilament light chain elevation and disability progression in multiple sclerosis. JAMA Neurol. 2023;80(12):1317–1325. doi: 10.1001/jamaneurol.2023.3997
[8] Domingues RB, Fernandes GBP, Leite F, et al. Neurofilament light chain in the assessment of patients with multiple sclerosis. Arq Neuro-Psiquiatr. 2019;77(6):436–441. doi: 10.1590/0004-282x20190060
[9] Puz P, Steposz A, Lasek-Bal A, et al. Diagnostic methods used in searching for markers of atrophy in patients with multiple sclerosis. Neurol Res. 2018;40(2):110–116. doi: 10.1080/01616412.2017.1403729
[10] Chatterjee P, Pedrini S, Doecke JD, et al. Plasma Aβ42/40 ratio, p-tau181, GFAP, and NfL across the Alzheimer's disease continuum: a cross-sectional and longitudinal study in the AIBL cohort. Alzheimers Dement. 2023;19(4):1117–1134. doi: 10.1002/alz.12724
[11] Smirnov DS, Ashton NJ, Blennow K, et al. Plasma biomarkers for Alzheimer's disease in relation to neuropathology and cognitive change. Acta Neuropathol. 2022;143(4):487–503. doi: 10.1007/s00401-022-02408-5
[12] Poesen K, De Schaepdryver M, Stubendorff B, et al. Neurofilament markers for ALS correlate with extent of upper and lower motor neuron disease. Neurology. 2017;88(24):2302–2309. doi: 10.1212/WNL.0000000000004029
[13] Poesen K, Van Damme P. Diagnostic and prognostic performance of neurofilaments in ALS. Front Neurol. 2018;9:1167. doi: 10.3389/fneur.2018.01167
[14] Hicks C, Dhiman A, Barrymore C, et al. Traumatic brain injury biomarkers, simulations and kinetics. Bioengineering (Basel). 2022;9(11):612. doi: 10.3390/bioengineering9110612
[15] Halbgebauer R, Halbgebauer S, Oeckl P, et al. Neurochemical monitoring of traumatic brain injury by the combined analysis of plasma Beta-Synuclein, NfL, and GFAP in polytraumatized patients. Int J Mol Sci. 2022;23(17):9639. doi: 10.3390/ijms23179639
[16] Schaller-Paule MA, Maiworm M, Schäfer JH, et al. Matching proposed clinical and MRI criteria of aggressive multiple sclerosis to serum and cerebrospinal fluid markers of neuroaxonal and glial injury. J Neurol. 2024;271(6):3512–3526. doi: 10.1007/s00415-024-12299-z
[17] Sotirchos ES, Fitzgerald KC, Singh CM, et al. Associations of sNfL with clinico-radiological measures in a large MS population. Ann Clin Transl Neurol. 2023;10(1):84–97. doi: 10.1002/acn3.51704
[18] Leuzy A, Mattsson-Carlgren N, Palmqvist S, et al. Blood-based biomarkers for Alzheimer's disease. EMBO Mol Med. 2022;14(1):e14408. doi: 10.15252/emmm.202114408
[19] Honig LS, Kang MS, Lee AJ, et al. Evaluation of plasma biomarkers for A/T/N classification of Alzheimer disease among adults of Caribbean Hispanic ethnicity. JAMA Netw Open. 2023;6(4):e238214. doi: 10.1001/jamanetworkopen.2023.8214
[20] Zanella I, Blasco H, Filosto M, et al. Editorial: the impact of Neurofilament Light chain (NFL) quantification in serum and cerebrospinal fluid in neurodegenerative diseases. Front Neurosci. 2022;16:915115. doi: 10.3389/fnins.2022.915115
[21] Coppens S, Lehmann S, Hopley C, et al. Neurofilament-light, a promising biomarker:

analytical, metrological and clinical challenges. Int J Mol Sci. 2023;24(14):11624. doi: 10.3390/ijms241411624
[22] Delaby C, Bousiges O, Bouvier D, et al. Neurofilaments contribution in clinic: state of the art. Front Aging Neurosci. 2022;14:1034684. doi: 10.3389/fnagi.2022.1034684
[23] Vrillon A, Ashton NJ, Karikari TK, et al. Comparison of CSF and plasma NfL and pNfH for Alzheimer's disease diagnosis: a memory clinic study. J Neurol. 2024;271(3):1297–1310. doi: 10.1007/s00415-023-12066-6
[24] Allen RP, Picchietti DL, Garcia-Borreguero D, et al. Restless legs syndrome/Willis–Ekbom disease diagnostic criteria: updated international restless legs syndrome study group (IRLSSG) consensus criteria–history, rationale, description, and significance. Sleep Med. 2014;15(8):860–873. doi: 10.1016/j.sleep.2014.03.025
[25] Hening W, Walters A, Rosen R, et al. The international RLS study group rating scale: a reliable and valid instrument for assessing severity of the restless legs syndrome. Neurology. Lippincott Williams & Wilkins 530 Walnut ST, 2001. p. 19106–3621, Philadelphia (PA) USA.
[26] Winkelmann J, Czamara D, Schormair B, et al. Genome-wide association study identifies novel restless legs syndrome susceptibility loci on 2p14 and 16q12. 1. PLOS Genet. 2011;7(7):e1002171. doi: 10.1371/journal.pgen.1002171
[27] Schormair B, Kemlink D, Roeske D, et al. PTPRD (protein tyrosine phosphatase receptor type delta) is associated with restless legs syndrome. Nat Genet. 2008;40(8):946–948. doi: 10.1038/ng.190
[28] Chen S, Ondo WG, Rao S, et al. Genomewide linkage scan identifies a novel susceptibility locus for restless legs syndrome on chromosome 9p. Am J Hum Genet. 2004;74(5):876–885. doi: 10.1086/420772
[29] Schormair B, Zhao C, Bell S, et al. Identification of novel risk loci for restless legs syndrome in genome-wide association studies in individuals of European ancestry: a meta-analysis. Lancet Neurol. 2017;16(11):898–907. doi: 10.1016/S1474-4422(17)30327-7
[30] Vlasie A, Trifu SC, Lupuleac C, et al. Restless legs syndrome: an overview of pathophysiology, comorbidities and therapeutic approaches (review). Exp Ther Med. 2022;23(2):185. doi: 10.3892/etm.2021.11108
[31] Schmidauer C, Sojer M, Seppi K, et al. Transcranial ultrasound shows nigral hypoechogenicity in restless legs syndrome. Ann Neurol Off J Am Neurological Assoc The Child Neurol Soc. 2005;58(4):630–634. doi: 10.1002/ana.20572
[32] Rizzo G, Li X, Galantucci S, et al. Brain imaging and networks in restless legs syndrome. Sleep Med. 2017;31:39–48. doi: 10.1016/j.sleep.2016.07.018
[33] Allen RP. Restless leg Syndrome/Willis-ekbom disease pathophysiology. Sleep Med Clin. 2015;10(3):207–214. doi: 10.1016/j.jsmc.2015.05.022
[34] Earley CJ, Kuwabara H, Wong DF, et al. Increased synaptic dopamine in the putamen in restless legs syndrome. Sleep. 2013;36(1):51–57. doi: 10.5665/sleep.2300
[35] Yin P, Niu X, Guan C, et al. Relationship between increased serum neurofilament light chain and glial fibrillary acidic protein levels with non-motor symptoms in patients with Parkinson's disease. Psychogeriatrics. 2024;24(2):415–425. doi: 10.1111/psyg.13088
[36] Mattsson N, Cullen NC, Andreasson U, et al. Association between longitudinal plasma neurofilament light and neurodegeneration in patients with Alzheimer disease. JAMA Neurol. 2019;76(7):791–799. doi: 10.1001/jamaneurol.2019.0765
[37] Diekämper E, Brix B, Stöcker W, et al. Neurofilament levels are reflecting the loss of presynaptic dopamine receptors in movement disorders. Front Neurosci. 2021;15:690013. doi: 10.3389/fnins.2021.690013
[38] Teunissen C, Iacobaeus E, Khademi M, et al. Combination of CSF N-acetylaspartate and neurofilaments in multiple sclerosis. Neurology. 2009;72(15):1322–1329. doi: 10.1212/WNL.0b013e3181a0fe3f
[39] Mariotto S, Savoldi A, Donadello K, et al. Nervous system: subclinical target of SARS-CoV-2 infection. J Neurol Neurosurg Psychiatry. 2020;91(9):1010–1012. doi: 10.1136/jnnp-2020-323881
[40] Virhammar J, Nääs A, Fällmar D, et al. Biomarkers for central nervous system injury in cerebrospinal fluid are elevated in COVID-19 and associated with neurological symptoms and disease severity. Eur J Neurol. 2021;28(10):3324–3331. doi: 10.1111/ene.14703
[41] Sahin BE, Celikbilek A, Kocak Y, et al. Neurological symptoms and neuronal damage markers in acute COVID-19: Is there a correlation? A pilot study. J Med Virol. 2023;95(1):e28240. doi: 10.1002/jmv.28240
[42] Gülçiçek A, Şanlı E, Şenel GB, et al. Patients with restless legs syndrome exhibit reduced serum colony-stimulating factor-1, humanin-like 3 and 10 levels. Acta Neurol Belg. 2024;124(5):1561–1568. doi: 10.1007/s13760-024-02549-2
[43] Kısabay A, Sarı US, Korkmaz T, et al. Evaluation of neurodegeneration through visual evoked potentials in restless legs syndrome. Acta Neurol Belg. 2016;116(4):605–613. doi: 10.1007/s13760-016-0631-4
[44] Truffi M, Garofalo M, Ricciardi A, et al. Neurofilament-light chain quantification by Simoa and Ella in plasma from patients with dementia: a comparative study. Sci Rep. 2023;13(1):4041. doi: 10.1038/s41598-023-29704-8