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Patterns of TDP-43 Deposition in Brains with LRRK2 G2019S Mutations

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Abstract

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OBJECTIVE—To assess for TDP-43 deposits in brains with and without a *LRRK2* G2019S mutation.

BACKGROUND—*LRRK2* G2019S mutations have been associated with parkinsonism and a wide range of pathological findings. There are no systematic studies examining the frequency and extent of TDP-43 deposits in neuropathological samples from *LRRK2* G2019S carriers.

METHODS—Twelve brains with *LRRK2* G2019S mutations were available for study from the New York Brain Bank at Columbia University; 11 of those had samples available for TDP-43 immunostaining. Clinical, demographic and pathological data are reported for 11 brains with a *LRRK2* G2019S mutation and compared to 11 brains without *GBA1* or *LRRK2* G2019S mutation with a pathologic diagnosis of PD or DLBD. They were frequency matched by age, gender, parkinsonism age of onset and disease duration.

RESULTS—TDP-43 aggregates were present in 73% (n=8) of brains with a *LRRK2* mutation and 18% (n=2) of brains without a *LRRK2* mutation (p=0.03). In one brain with a *LRRK2* mutation, TDP-43 proteinopathy was the primary neuropathological change.

CONCLUSIONS—Extra-nuclear TDP-43 aggregates are observed with greater frequency in *LRRK2* G2019S autopsies compared to PD cases without a *LRRK2* G2019S mutation. The association between *LRRK2* and TDP-43 should be further explored.

INTRODUCTION

The *LRRK2* G2019S mutation is present in 1–4% of all Parkinson's disease (PD) cases and is more common in selected populations, namely North African Berbers (37% in familial cases and 41% in sporadic cases) and Ashkenazi Jews (23% in familial cases and 13% in sporadic cases) (1–4). Penetrance estimates are variable ranging from 25%−100% (3–5). A key striking feature of *LRRK2*-associated PD is the variability of neuropathologic findings even from related individuals, especially with regards to the presence of Lewy bodies (LB). When examined for other neuropathologic findings, some *LRRK2* cases without alpha-synuclein or phosphorylated tau deposition have been found to have transactivation-response DNA binding protein of 43kDa (TDP-43) containing cytoplasmic inclusions (6–8).

TDP-43, which is a RNA/DNA binding intranuclear protein that regulates mRNA splicing, translation, transportation and degradation (9), is found mislocalized in the cytoplasm in the form of inclusions in many neurodegenerative diseases (10–15). TDP-43 was first found in ubiquitin positive inclusions in the central nervous system in cases of amyotrophic lateral sclerosis (ALS) and frontotemporal lobar dementia (FTLD) (9, 16). Subsequently, cytoplasmic TDP-43-positive inclusions were also found to be a key neuropathological features of the newly recognized neuropathological entity called limbic predominant age related TDP-43 encephalopathy neuropathological change (LATE-NC), which is frequently seen in Lewy body disorders (LBD) and Alzheimer's disease (AD) (17).

The frequency of TDP-43-associated pathology in *LRRK2* PD cases is unclear since most autopsy reports have not included TDP-43 immunostaining. In the literature, five cases with *LRRK2* pathogenic variants are described as having TDP-43-positive cytoplasmic inclusions and neurites (7, 8, 18, 19).

In this case series, we examined our collection of *LRRK2* brains to better understand the frequency of TDP-43 pathology in cases with a *LRRK2* G2019S mutation. We compared brains with a *LRRK2* mutation to brains with a pathologic diagnosis of PD or diffuse Lewy body disease (DLBD) without *LRRK2* or *GBA1* mutations to see if TDP-43 pathology is unique to *LRRK2* carriers.

METHODS

Materials and methods

All brains were processed using the standardized protocol as outlined previously (20). In brief, fresh brains were divided by a sagittal cut through the corpus callosum and brainstem; one half brain is processed for banking of fresh-frozen samples for research and the contralateral half is immersed in formalin and processed for neuropathologic evaluation. Determination of PD and Braak neuropathologic stage was performed according to published criteria (21–23).

Histochemical staining protocols

In addition to sections stained with Luxol-fast blue and counterstained with hematoxylin and eosin, sections from selected blocks were separately immunostained for alpha-synuclein and TDP-43 (supplementary table 1) using an automated immunostaining platform that utilizes a 3,3’-diaminobenzidine-based immunostaining method. Hematoxylin counterstains were performed.

TDP-43 immunohistochemical stains were performed on sections of the amygdala, hippocampal formation at the level of the lateral geniculate body and superior frontal cortex (Brodmann area 9) for determination of LATE-NC staging (24). Additionally, TDP-43 immunohistochemical stains were performed on the midbrain.

Clinical information and case selection—All brains were screened as previously described for the *LRRK2* G2019S and 10 *GBA1* mutations and variants (25, 26). Of the twelve cases available with a *LRRK2* G2019S mutation, eleven cases had samples available with TDP-43 staining. The cases without a *LRRK2* or *GBA1* mutation were selected if they had a pathologic diagnosis of PD or DLBD; cases were selected in a blinded fashion regarding TDP-43 pathology. Cases with and without a *LRRK2* mutation were matched to the best of our ability by gender, age of onset and disease duration. Clinical information was obtained by chart review of medical records from Columbia University and outside records, if applicable. All cases were clinically diagnosed with PD by a movement disorders neurologist. Demographics were recorded for each patient reviewed.

Statistical analysis—To determine the significance of TDP-43 deposition in cases with and without a *LRRK2* G2019S mutation, Fisher Exact analysis was performed using SPSS version 28.0.

RESULTS

The demographics and clinical characteristics of all the cases reviewed are presented in Table 1. By design, there was no difference between the groups in age, gender and disease duration.

TDP-43 extra-nuclear deposits were present in 8 of 11 brains (73%) with a *LRRK2* mutation and in 2 of 11 (18%) non-carrier PD brains (p=0.03, Fisher Exact). The deposits in the *LRRK2* brains were seen in three different patterns:

1. TDP-43 dystrophic neurites (DN) in the amygdala were present in all positively stained brains (n = 8), and among them five exclusively in the amygdala (Figure 1A).
2. Two brains had TDP-43 DN in the substantia nigra (SN) (Figure 1B).
3. One patient had FTLD Type C with neuronal loss and gliosis primarily in the frontal lobe and aberrant TDP-43 staining mainly in the prefrontal, motor and temporal cortices. Neuronal cytoplasmic inclusions (NCI) were seen in the frontal lobe, hippocampus and amygdala (Figure 1C).

For the non-carrier brains, one brain had NCI in the hippocampus and the amygdala. Another brain had DN in the frontal cortex, hippocampus and amygdala. None of the non-carrier brains had TDP-43 deposition in the midbrain.

A summary of the distribution of TDP-43, alpha-synuclein, tau and amyloid deposits for cases with a *LRRK2* G2019S mutation are summarized in Supplementary Table 2 and non-carrier brains are summarized in Supplementary Table 3.

The patient with FTLD type C clinically had levodopa responsive parkinsonism. This patient had an 18-year clinical course with initial symptoms of slowness of movement and cognitive changes. She was clinically diagnosed with PD and had a significant response to levodopa with motor fluctuations addressed with a dopamine agonist. She had a rest tremor in her left leg, and ultimately developed motor fluctuations with sudden offs that manifested as anxiety, panic, urinary urgency, rigidity and pain. Clinical histories for the *LRRK2* cases are summarized in the supplementary material section.

DISCUSSION

In this case series, TDP-43 pathology was assessed in 22 postmortem brains from patients with a clinical diagnosis of parkinsonism and we found that TDP-43 pathology is more common in brains with a *LRRK2* mutation. TDP-43 deposits were more common in *LRRK2* G2019S brains (73% versus 18%, p= 0.03, Fisher Exact), and among the *LRRK2* brains TDP-43 deposits were heterogenous. Specifically, one case had TDP-43 as the primary pathology (FTLD type C), all had DN in the amygdala and two cases had DN in the SN without any NCI. For the cases without a *LRRK2* mutation, one case had DN in the amygdala and hippocampus and the other case had NCI in the frontal cortex.

The phenotype of TDP-43 pathology in the SN in the absence of LB in *LRRK2* PD may be parkinsonism. The two *LRRK2* cases reported in the literature with these pathologic features, and case 2 from our series, had levodopa responsive parkinsonism with resting tremor, bradykinesia and rigidity (6, 19). These patients are clinically indistinguishable from idiopathic PD. None of our cases without a *LRRK2* mutation had TDP-43 pathology in the SN. In the literature, one case without any genetic mutations and a clinical history of PD was reported to exclusively have TDP-43 proteinopathy and neuronal loss in the SN (27).

The frequency of TDP-43 pathology reported in PD is variable and is not routinely screened for on autopsy. 18% of our cases without a *LRRK2* mutation had TDP-43 pathology. This is similar to what has been reported in autopsy studies where TDP-43 pathology was seen in 7–24% of cases (14, 28). To our knowledge this is the first study to look at the frequency in cases with a *LRRK2* mutation where a much higher percentage of cases (73%) had TDP-43 aggregates. Only one additional case in the literature has been described of a patient with FTLD and a *LRRK2* G2019S mutation(29). Clinically, the patient had postural and action tremors, but no signs of parkinsonism. It is unclear why these cases had more incident TDP-43 pathology, but *LRRK2* mutations may play a role in TDP-43 aggregation.

In conclusion, this is the largest series to date investigating the association between *LRRK2* mutations and TDP-43 aggregation. While all participants in the current series were clinically diagnosed with PD, it is estimated that most carriers of the G2019S mutation will never develop PD(3). The heterogeneity of the pathology in brains with *LRRK2* mutations may raise the question of the causality of the mutation in the neurodegenerative process; however, the significant increase in frequency of TDP-43 aggregates in *LRRK2* cases and the presence of tau in all our cases (regardless of *LRRK2* mutation status) supports the causative role it may play specifically for TDP-43 aggregates. Although the other co-pathologies present in all our cases make it challenging to propose a causal role for TDP-43 pathology, it is tempting to postulate that TDP-43 might be pathogenic in our case with FTLD type C where it is the primary pathology. The small size of our cohort with limited clinical history prevents us from making conclusions regarding the clinical phenotype of TDP-43 proteinopathy. Our genetic data is limited to *LRRK2* and *GBA1* and future work will look into other genetic markers such as *TMEM106B* and *GRN* for our FTLD case and *APOE* for the cases with AD pathology. In this case series, we were limited by having only non-phosphorylated TDP-43 analysis. Given that *LRRK2* G2019S is associated with increased kinase activity(30) and given the many phosphorylation sites on TDP-43(31) future studies should compare the level of phosphorylation of TDP-43 in *LRRK2* G2019S brains to idiopathic PD. We also suspect that there may be seeding between TDP-43 and synuclein. Colocalization was observed in the autopsy series reported by Uemura and colleagues (17) and future studies would use double immunostaining to evaluate for this in *LRRK2* brains. The presence of TDP-43 in LBD underscores the need to screen for it in postmortem brains of patients who have a history of parkinsonism, especially if they have a *LRRK2* mutation. Further investigation is needed to better understand the potential biological link between *LRRK2* mutations and TDP-43 aggregation.

Supplementary Material

Refer to Web version on PubMed Central for supplementary material.

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References

1. Healy DG, Falchi M, O'Sullivan SS, Bonifati V, Durr A, Bressman S, et al. Phenotype, genotype, and worldwide genetic penetrance of LRRK2-associated Parkinson's disease: a case-control study. Lancet Neurol. 2008;7(7):583–90. [PubMed: 18539534]
2. Schulte C, Gasser T. Genetic basis of Parkinson's disease: inheritance, penetrance, and expression. Appl Clin Genet. 2011;4:67–80. [PubMed: 23776368]
3. Lee AJ, Wang Y, Alcalay RN, Mejia-Santana H, Saunders-Pullman R, Bressman S, et al. Penetrance estimate of LRRK2 p.G2019S mutation in individuals of non-Ashkenazi Jewish ancestry. Mov Disord. 2017;32(10):1432–8. [PubMed: 28639421]
4. Lesage S, Ibanez P, Lohmann E, Pollak P, Tison F, Tazir M, et al. G2019S LRRK2 mutation in French and North African families with Parkinson's disease. Ann Neurol. 2005;58(5):784–7. [PubMed: 16240353]
5. Marder K, Wang Y, Alcalay RN, Mejia-Santana H, Tang MX, Lee A, et al. Age-specific penetrance of LRRK2 G2019S in the Michael J. Fox Ashkenazi Jewish LRRK2 Consortium. Neurology. 2015;85(1):89–95. [PubMed: 26062626]
6. Zimprich A, Biskup S, Leitner P, Lichtner P, Farrer M, Lincoln S, et al. Mutations in LRRK2 cause autosomal-dominant parkinsonism with pleomorphic pathology. Neuron. 2004;44(4):601–7. [PubMed: 15541309]
7. Wider C, Dickson DW, Wszolek ZK. Leucine-rich repeat kinase 2 gene-associated disease: redefining genotype-phenotype correlation. Neurodegener Dis. 2010;7(1–3):175–9. [PubMed: 20197701]
8. Ling H, Kara E, Bandopadhyay R, Hardy J, Holton J, Xiromerisiou G, et al. TDP-43 pathology in a patient carrying G2019S LRRK2 mutation and a novel p.Q124E MAPT. Neurobiol Aging. 2013;34(12):2889 e5–9.
9. Gao J, Wang L, Huntley ML, Perry G, Wang X. Pathomechanisms of TDP-43 in neurodegeneration. J Neurochem. 2018.
10. Nelson PT, Dickson DW, Trojanowski JQ, Jack CR, Boyle PA, Arfanakis K, et al. Limbic-predominant age-related TDP-43 encephalopathy (LATE): consensus working group report. Brain. 2019;142(6):1503–27. [PubMed: 31039256]
11. Kwong LK, Neumann M, Sampathu DM, Lee VM, Trojanowski JQ. TDP-43 proteinopathy: the neuropathology underlying major forms of sporadic and familial frontotemporal lobar degeneration and motor neuron disease. Acta Neuropathol. 2007;114(1):63–70. [PubMed: 17492294]

12. Dewan R, Chia R, Ding J, Hickman RA, Stein TD, Abramzon Y, et al. Pathogenic Huntingtin Repeat Expansions in Patients with Frontotemporal Dementia and Amyotrophic Lateral Sclerosis. Neuron. 2021;109(3):448–60 e4. [PubMed: 33242422]
13. Hickman RA, Dewan R, Cortes E, Traynor BJ, Marder K, Vonsattel JP. Amyotrophic lateral sclerosis is over-represented in two Huntington's disease brain bank cohorts: further evidence to support genetic pleiotropy of pathogenic HTT gene expansion. Acta Neuropathol. 2022;143(1):105–8. [PubMed: 34800149]
14. Nakashima-Yasuda H, Uryu K, Robinson J, Xie SX, Hurtig H, Duda JE, et al. Co-morbidity of TDP-43 proteinopathy in Lewy body related diseases. Acta Neuropathol. 2007;114(3):221–9. [PubMed: 17653732]
15. Koga S, Lin WL, Walton RL, Ross OA, Dickson DW. TDP-43 pathology in multiple system atrophy: colocalization of TDP-43 and alpha-synuclein in glial cytoplasmic inclusions. Neuropathol Appl Neurobiol. 2018;44(7):707–21. [PubMed: 29660838]
16. Arai T, Hasegawa M, Akiyama H, Ikeda K, Nonaka T, Mori H, et al. TDP-43 is a component of ubiquitin-positive tau-negative inclusions in frontotemporal lobar degeneration and amyotrophic lateral sclerosis. Biochem Biophys Res Commun. 2006;351(3):602–11. [PubMed: 17084815]
17. Uemura MT, Robinson JL, Cousins KAQ, Tropea TF, Kargilis DC, McBride JD, et al. Distinct characteristics of limbic-predominant age-related TDP-43 encephalopathy in Lewy body disease. Acta Neuropathol. 2021.
18. Covy JP, Yuan W, Waxman EA, Hurtig HI, Van Deerlin VM, Giasson BI. Clinical and pathological characteristics of patients with leucine-rich repeat kinase-2 mutations. Mov Disord. 2009;24(1):32–9. [PubMed: 19006185]
19. Sakuwa M, Adachi T, Suzuki Y, Yoshida K, Fukuda H, Miura H, et al. First Japanese autopsy case showing LRRK2 mutation G2019S and TDP-43 proteinopathy. Parkinsonism Relat Disord. 2021;91:85–7. [PubMed: 34543853]
20. Vonsattel JP, Del Amaya MP, Keller CE. Twenty-first century brain banking. Processing brains for research: the Columbia University methods. Acta Neuropathol. 2008;115(5):509–32. [PubMed: 17985145]
21. Braak H, Del Tredici K, Rüb U, De Vos RA, Steur ENJ, Braak E. Staging of brain pathology related to sporadic Parkinson's disease. Neurobiology of aging. 2003;24(2):197–211. [PubMed: 12498954]
22. Gelb DJ, Oliver E, Gilman S. Diagnostic Criteria for Parkinson Disease. Archives of Neurology. 1999;56(1):33–9. [PubMed: 9923759]
23. Dickson DW. Neuropathology of Parkinson disease. Parkinsonism & related disorders. 2018;46:S30–S3. [PubMed: 28780180]
24. Nelson PT, Dickson DW, Trojanowski JQ, Jack CR, Boyle PA, Arfanakis K, et al. Limbic-predominant age-related TDP-43 encephalopathy (LATE): consensus working group report. Brain. 2019;142(6):1503–27. [PubMed: 31039256]
25. Alcalay RN, Dinur T, Quinn T, Sakanaka K, Levy O, Waters C, et al. Comparison of Parkinson risk in Ashkenazi Jewish patients with Gaucher disease and GBA heterozygotes. JAMA Neurol. 2014;71(6):752–7. [PubMed: 24756352]
26. Alcalay RN, Mirelman A, Saunders-Pullman R, Tang MX, Mejia Santana H, Raymond D, et al. Parkinson disease phenotype in Ashkenazi Jews with and without LRRK2 G2019S mutations. Mov Disord. 2013;28(14):1966–71. [PubMed: 24243757]
27. Yamashita R, Beck G, Yonenobu Y, Inoue K, Mitsutake A, Ishiura H, et al. TDP-43 Proteinopathy Presenting with Typical Symptoms of Parkinson's Disease. Mov Disord. 2022;37(7):1561–3. [PubMed: 35531755]
28. Yokota O, Davidson Y, Arai T, Hasegawa M, Akiyama H, Ishizu H, et al. Effect of topographical distribution of alpha-synuclein pathology on TDP-43 accumulation in Lewy body disease. Acta Neuropathol. 2010;120(6):789–801. [PubMed: 20669025]
29. Dachsel JC, Ross OA, Mata IF, Kachergus J, Toft M, Cannon A, et al. Lrrk2 G2019S substitution in frontotemporal lobar degeneration with ubiquitin-immunoreactive neuronal inclusions. Acta Neuropathol. 2007;113(5):601–6. [PubMed: 17151837]

30. Kalogeropulou AF, Purlyte E, Tonelli F, Lange SM, Wightman M, Prescott AR, et al. Impact of 100 LRRK2 variants linked to Parkinson's disease on kinase activity and microtubule binding. Biochem J. 2022;479(17):1759–83. [PubMed: 35950872]

31. Eck RJ, Kraemer BC, Liachko NF. Regulation of TDP-43 phosphorylation in aging and disease. Geroscience. 2021;43(4):1605–14. [PubMed: 34032984]

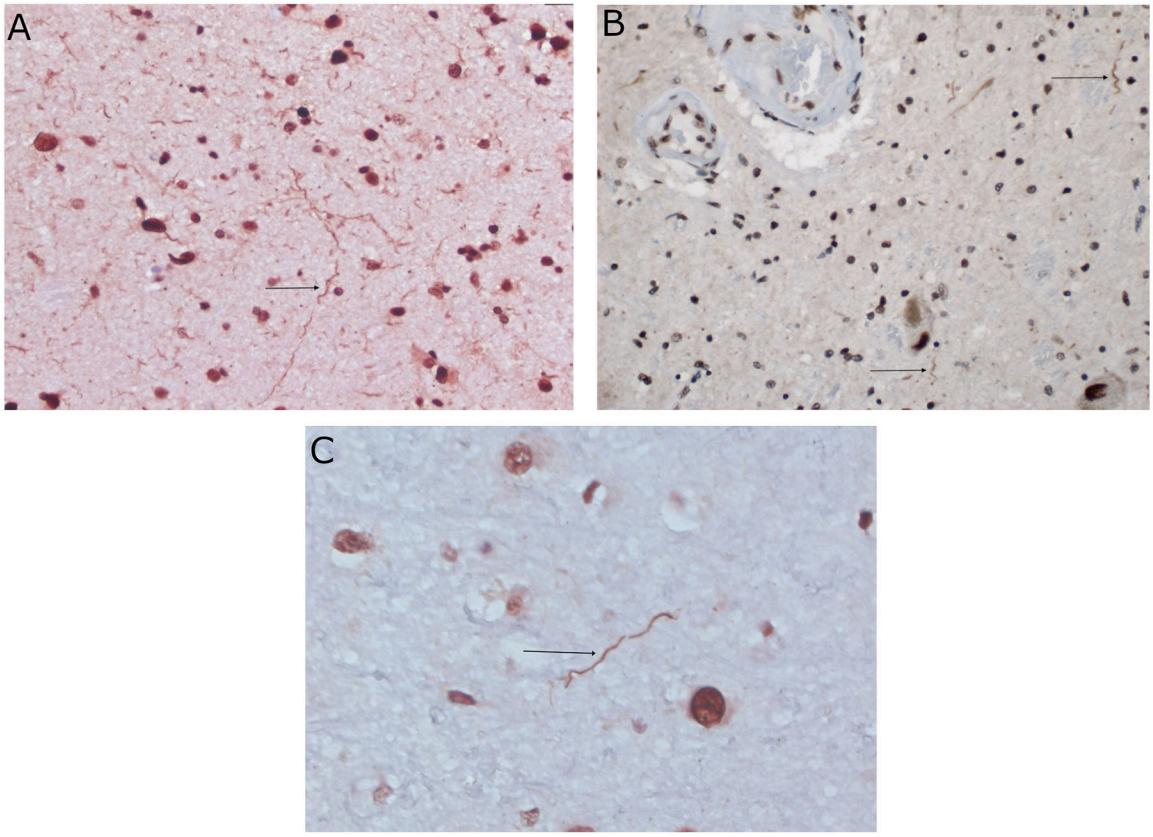

Figure 1 Legend:
Photomicrographs of immunohistochemical stains against non-phosphorylated TDP-43. A: Arrow pointing to dystrophic neurites in amygdala of case 8. Original magnification 400X; TDP-43. B: Arrows pointing to dystrophic neurites in substantia nigra of case 1. Original magnification 400X; TDP-43. C: Arrow pointing to a long, thick dystrophic neurite in the motor cortex of case 7 with a pathologic diagnosis of FTLD type C. Original magnification 630X; TDP-43.

Table 1:

Demographics

	Cases with *LRRK2* G2019S mutation	**PD cases without *LRRK2* or *GBA1* mutations**
Number of cases (n)	11	11
Female gender % (n)	36% (4)	45% (5)
Parkinsonism age of onset (mean; years-of-age)	61 ± 11.2	56.9 ± 9.4
Mean disease duration (mean; years ± SD)	18.6 ± 7.5	20.8 ± 6.9
Age of death (mean; years of age ± SD)	79.3 ± 8.6	77.6 ± 8