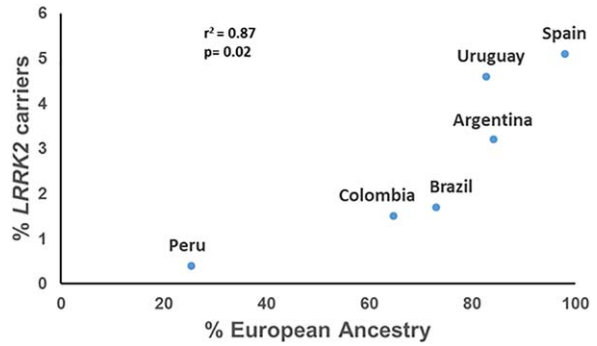

FIG. 1. Correlation between the frequency of *LRRK2* carriers and the estimated proportion of European ancestry by population. The percentage of *LRRK2* carriers represents the combined number of PD patients carrying *LRRK2*-p.G2019S and *LRRK2*-p.R1441G/C/H/S per population: Argentina (6 of 188); Brazil (7 of 433); Colombia (3 of 197); Peru (2 of 543); and Uruguay (13 of 288). The combined *LRRK2* mutation frequency derived for three cohorts from Spain (67 of 1,319)[6-8] is provided for reference. Correlation (coefficient of determination, r^2) and significance level (*P* value) are also shown. [Color figure can be viewed at wileyonlinelibrary.com]

PD case-control sample of Latinos, has been accomplished, and to date nearly 4,000 individuals have been enrolled.

Mutations located in codons G2019 and R1441 of the leucine-rich repeat kinase 2 (*LRRK2*) gene represent the most common genetic cause of PD in patients of European origin. Our group and others have shown that these mutations also occur in PD patients from Latin America, where most carriers share haplotypes that have been widely reported in European populations.[3,4] Here, we expand on our previous work[4] to show that, in the LARGE-PD cohort, the combined frequency of these *LRRK2* mutations varies substantially across countries and is directly correlated with the estimated proportion of European ancestry at each site (Fig. 1). This indicates that the genetic architecture of PD might differ between Latinos and other population groups, and this same observation has been made in several other diseases, including cancer.[5] With a new grant from the PDF, we are now embarking on large-scale analyses to more thoroughly characterize the genetics of PD in LARGE-PD. Because replication in such endeavors is critical, we have begun to recruit an independent Latino PD case-control sample in the United States in collaboration with several movement disorder centers across the country.

In an era where our field is actively seeking more individualized treatments (precision medicine), it is critical that we avoid continuation of the Eurocentric model of genetic research. We believe that LARGE-PD represents a much needed break with the status quo. ■

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Appendix

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Neuromelanin Magnetic Resonance Imaging of the Substantia Nigra in *LRRK2*-Related Parkinson's Disease

Specific T_1-weighted MRI sequences are able to detect SN neuromelanin (NM) signal changes and accurately discriminate Parkinson's disease (PD) patients from controls.[1,2]

The study of NM-MRI in PD patients carrying a *LRRK2* gene mutation (*LRRK2*-PD) could contribute to further uncover *LRRK2*-associated phenotype. Albeit considered to largely overlap idiopathic PD (iPD),[3] differences have been described.[4] Furthermore, the identification of a biomarker of

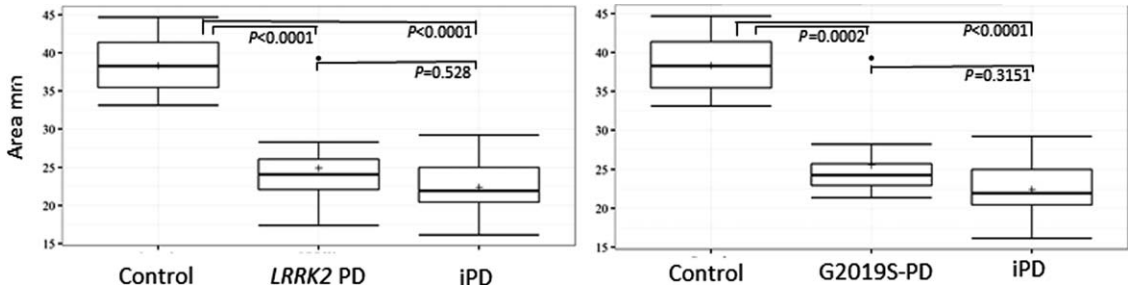

FIG. 1. Area of the SN in LRRK2-PD, G2019S-PD, iPD and control individuals. *LRRK2*-PD: all PD patients carrying a *LRRK2* mutation (G2019S and R1441H); G2019S-PD: PD patients carrying a G2019S mutation.

neurodegeneration in *LRRK2*-PD can eventually support studies in asymptomatic carriers. Castellanos and colleagues[5] found NM-MRI SN volumes significantly reduced in both idiopathic and *LRRK2*-PD (3 G2019S and 4 R1441G PD patients).

Our study aimed to further investigate neuromelanin imaging in *LRRK2*-PD. We performed a cross-sectional study including *LRRK2*-PD patients, control individuals with no signs or family history of a neurodegenerative disorder, and PD patients with no *LRRK2* mutations identified (referred as iPD). Our primary outcome was SN neuromelanin high signal area obtained with semiautomated methods.

LRRK2-PD and iPD patients were identified through previous[6] and recent genetic studies. Clinical assessments were performed in best *On*. Imaging was acquired using a 3.0 Tesla scanner and NM-sensitive pulse sequence was used as previously described.[1] OsiriX software[7] was used for imaging postprocessing. Data analysis was blinded to clinical and genetic status. Kruskal-Wallis, with pair-wise comparisons (Bonferroni method applied), and Mann-Whitney U tests were used as appropriate ($P < 0.05$). Nonparametric receiver operating characteristic curves were constructed for calculating NM imaging area sensitivity and specificity for discriminating groups.

Thirteen *LRRK2*-PD patients (10 G2019S, 3 R1441H), 10 controls, and 13 iPD patients were included. No significant differences between groups were identified concerning sex, age at disease onset (59.7 ± 12.3 *LRRK2*-PD vs. 61.8 ± 11.8 iPD), disease duration (7.7 ± 3.5 *LRRK2*-PD vs. 10.7 ± 4.3 iPD), MDS-UPDRS I, III (36.8 ± 13.0 *LRRK2*-PD; 41.2 ± 16.2 iPD), and IV scores, or levodopa equivalent daily dose. Mean age at examination in *LRRK2*-PD (67.4 ± 12.9), control (61.2 ± 7.4), and iPD (72.5 ± 12.7) groups were different. Although when comparing *LRRK2*-PD versus controls ($P = 0.1640$) and *LRRK2*-PD versus iPD ($P = 0.3220$) mean age at examination was not statistically significantly different, iPD group presented a significantly higher mean age at examination when compared to controls ($P = 0.0211$), limiting results interpretation. The H & Y (2.0 ± 0.1 *LRRK2*-PD vs. 3.0 ± 0.9 iPD) and MDS-UPDRS II scores were significantly worse in iPD.

Median SN NM area was significantly decreased in the *LRRK2*-PD group compared to controls (Fig. 1). Furthermore, when only considering G2019S *LRRK2*-PD, median SN NM signal area was also significantly decreased compared to controls. No differences were found between *LRRK2*-PD and iPD groups. High signal area showed 92.3% sensitivity and 100% specificity for discriminating *LRRK2*-PD patients from controls (cut off: 28.24 mm^2).

In our study, NM-MR imaging of the SN was able to differentiate *LRRK2*-PD patients from controls, and NM signal area reductions in *LRRK2*-PD and iPD groups did not show statistically significant differences. Our results present the most extensive data on NM-MRI in *LRRK2*-related PD and support NM imaging as a potential biomarker of SN degeneration in *LRRK2*-related PD. ■

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Pathophysiological Heterogeneity in Parkinson's Disease: Neurophysiological Insights from LRRK2 Mutations

Mutations in the leucine-rich repeat kinase 2 (*LRRK2*) are the most common cause of hereditary Parkinson's disease (PD).[1] Although the phenotype of PD patients with and without LRRK2 G2019S mutations is similar, some differences in clinical characteristics have been described in large clinical series,[2] suggesting that there may be differences in pathophysiology. A recent neurophysiological study suggested that LRRK2 patients have impaired GABAa inhibition in the cerebral cortex.[3] Here, we report additional neurophysiological data on 6 unrelated PD patients with G2019S LRRK2 mutation (LRRK2 PD) and compared it with data from 12 sporadic PD patients and 15 healthy participants (HP). The studies were carried out after overnight withdrawal of levodopa and examined function in the less-affected hemisphere. The local ethics committee approved the study and written informed consent was obtained.

Patients' demographic and clinical data are given in Supporting Table 1. There were no differences in age, sex distribution, disease duration, or UPDRS motor scores between groups. Transcranial magnetic stimulation (TMS) coupled with surface electromyography recordings of motor evoked potentials (MEPs) in the abductor pollicis brevis muscle (APB) were used to measure: resting and active motor thresholds (RMT, AMT); input/output (I/O) curve; short interval intracortical inhibition (SICI) at interstimulus intervals (ISI) of 2, 3, and 4 ms using a conditioning pulse intensity of 90% RMT; intracortical facilitation (ICF; ISI = 15 ms); cortical silent period (intensity 120% RMT); long interval intracortical inhibition (LICI; ISI = 100, 150, and 200 ms, conditioning pulse intensity 150% RMT), using the same methods as previously described.[4] Plasticity was probed using the standard excitatory paired associative stimulation (PAS) protocol.[4]

The results with statistics are illustrated in Figure 1. LRRK2 PDs had steeper I/O curve and less effective SICI compared to sporadic PDs. CSP and LICI were reduced in LRRK2 patients compared to HP, but not different from sporadic PDs. No difference in PAS response was found between groups.

Our study confirms that LRRK2 PDs compared to sporadic PDs have less-effective SICI,[3] a measure of GABAa activity,[5] suggesting that SICI may be a useful electrophysiological measure to distinguish patients with and without LRRK2 mutation at a group level. Although our LRRK2 patients showed reduced CSP and LICI compared to HC, which reflect GABAb activity,[5] these were not able to differentiate LRRK2 from sporadic PDs. In addition, we describe increased corticospinal excitability in LRRK2 PDs compared to sporadic PDs, as evident in increased steepness of I/O curve. There was no correlation between UPDRS scores and any of the TMS measures in PD patients, suggesting that electrophysiological differences between PD groups are unlikely to be the result of small differences in severity of motor impairment.

Changes in TMS measures in LRRK2 have been attributed to reduced GABAergic inhibition,[3] and our data on reduced SICI confirm this hypothesis. One possibility is that there is also an increased excitability of glutamatergic systems in LRRK2,[6] suggested by the increased steepness of the I/O curve observed in our LRRK2 patients. To check the hypotheses of disruption of the normal balance between intracortical inhibition and excitation, experiments on a large number of patients is needed. Considering the rarity of LRRK2 mutation, we call for a multicentric study. ■

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