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Prospective clinical and DaT-SPECT imaging in premotor *LRRK2* G2019S-associated Parkinson disease

~~Progression to Parkinson's disease in carriers of *LRRK2* G2019S mutation: a 4-year prospective study with serial dopamine transporter imaging~~

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Abstract

Objective: To asses the value of baseline clinical and imaging biomarkers in a cohort of asymptomatic *LRRK2* G2019S carriers for predicting conversion to Parkinson's disease (PD) at 4-years.

Methods: Thirty-two asymptomatic carriers of *LRRK2* G2019S mutation, with baseline and 4-year evaluation including clinical examination (UPDRS-III, olfaction UPSIT test) and DaT-SPECT (^{123}I-ioflupane). Visual and semiquantitative analysis of images was performed. The specific striatal binding ratio was calculated (striatal ROI-occipital ROI/occipital ROI).

Results: Three carriers, asymptomatic at baseline, had converted to PD at 4-year evaluation. Twenty-three participants were fully evaluated. PD-converters had lower striatal DaT binding at baseline than non-converters (p= 0.002). A baseline scan with a ratio of bilateral striatal uptake below 1 predicted with high sensitivity and specificity conversion to PD within the 4-year period (AUC=1, p= 0.006). The slope of DaT

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binding decline between the two scans was similar in PD-converters and non-converters. Age-adjusted UPSIT score at baseline and at 4-years was similar in both groups.

Conclusions: Semiquantitative DaT-SPECT could be used to predict early conversion to PD in asymptomatic carriers of *LRRK2* G2019S mutation. Rate of conversion to PD at 4 years in this cohort aged ~ 64 years was 12%. The slope of DaT binding decline on DaT-SPECT imaging seems to be similar across different stages of the premotor period.

The G2019S mutation of the *LRRK2* gene is the commonest known cause of Parkinson's disease (PD) to date[1]. Clinically and pathologically this form of the disease is indistinguishable from idiopathic PD and therefore it constitutes an ideal scenario in which to deepen our knowledge about the disease. The identification and follow-up of carriers of the *LRRK2* G2019S mutation that still haven't developed motor symptoms of PD represents a unique opportunity for studying the prodromal stage of Parkinson's disease. We still lack answers for questions like how long before the appearance of the motor symptoms of PD the neurodegenerative process starts and what's the pace of cell loss at the *substantia nigra* (SN) at the premotor stage ~~of PD~~. Shed~~ee~~ding light into the prodromal stages of the disease is going to be crucial for planning drug trials aiming to modify the disease process early, before most of the damage is already done. The penetrance of the *LRRK2* G2019S mutation is age-dependent and only a proportion of *LRRK2* G2019S mutation carriers will develop motor symptoms of PD[2]. Besides age no

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other clinical marker has been proven to be useful to predict conversion to manifest PD in asymptomatic carriers. In a previous study of our group we characterized with brain imaging (^{123}I-ioflupane SPECT and transcranial sonography) and olfaction tests a cohort of 32 asymptomatic carriers of the *LRRK2* G2019S mutation[3]. Here we report the findings of a re-evaluation of this cohort at 4 years, including clinical examination, olfaction testing and ^{123}I-ioflupane SPECT. Our objective was double: first to assess whether any of the biomarkers studied at baseline was helpful to predict conversion to PD within~~in~~ the 4-year period, and second to estimate through sequential ^{123}I-ioflupane imaging the rate of progression of the dopaminergic terminal loss in the premotor stage of *LRRK2* G2019S-associated PD.

Methods

Patients

This cohort was composed of 32 asymptomatic carriers of *LRRK2* G2019S mutation reported in a previous study[3]. These were the participants~~subjects~~ from that study that had completed all the tests (clinical examination, transcranial sonograhy, ^{123}I-ioflupane SPECT and olfaction test). All of them were relatives of *LRRK2* G2019S PD proband cases identified from the study of 367 consecutive patients with PD attended at a single center~~, the University Hospital Marqués de Valdecilla in Santander, northern Spain~~[2]. Participants belonged to a total of 21 pedigrees.

Clinical assessment

Approximately four years after the baseline evaluation the 32 participants were invited

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telephonically to participate in the study. Those who accepted were examined for the presence of signs of PD. We used the Unified Parkinson's Disease Rating Scale, part III (UPDRS-III), and a diagnosis of PD was made according to the Queen Square Brain Bank Criteria[4]. All participants were assessed at a single center and by the same research team involved in the baseline evaluation, in which examiners were blind to the genetic status of *LRRK2* relatives[3]. Clinical examination was done before the rest of the tests were available so examiners were blind to these results. In cases in which the examination led to a diagnosis of PD we acceded to patient's clinical records to check if the patient had already been diagnosed of PD within the 4-year period.

Olfaction testing

Olfaction was evaluated through the encapsulated odor University of Pennsylvania Smell Identification Test (UPSIT)[5]. The test was self-administered by using standard 40-odor identification, either at the time of the visit or at the participant~~subject~~'s residence, and was returned by mail. Participants ~~Subjects~~ were asked to choose a response from the four choices listed. Tests including incomplete responses were excluded from the analysis. ~~Participants with a known respiratory tract infection or active allergies as well as those with previous conditions known to impair the sense of smell were excluded from this part of the study.~~

^{123}I-ioflupane SPECT protocol

Acquisition of ^{123}I-ioflupane SPECT images was performed as described elsewere[3]. ~~One hour prior to the injection of the radiotracer an oral solution of lugol was administered to blockade the thyroid uptake and to prevent exposure to free ^{123}I. Patients were~~

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~~injected with 148 MBq of ^{123}I ioflupane (DaTSCAN, GE Healthcare. Eindhoven, The Netherlands). Acquisition was obtained with patient in supine position. A head holder was used to minimize motion artifacts. Brain SPECT was performed in a dual detector gamma camera (E.CAM Siemens. Chicago, IL, USA) equipped with fan beam collimators. Images were acquired 3 hours after radiotracer injection, using the smallest possible rotational radius (<16 cm in all cases), 128 x 128 matrix, zoom factor 1, 25 seconds per position and 2 x 64 views.~~ For image processing filtered backprojection with a butterworth 0.55 filter and manual Chang attenuation correction with a coefficient of 0.12 cm-1 was applied. The axial slices were reoriented parallel to the frontal-occipital line. Visual and semiquantitative analysis of images was performed by two experienced nuclear medicine specialists blinded to clinical data (IMR~~JB~~ and MJA~~IMR~~). For the visual analysis images were categorized as negative or positive (when a decrease in the striatal uptake was observed). For the semiquantitative analysis, two region of interest (ROI) with the same size for all patients were manually drawn including the left and right striatum, respectively. These ROIs were placed in three consecutive axial slices showing the highest striatal uptake. The occipital cortex was selected as background to determine the nonspecific binding, using a rectangular ROI in the same three consecutive slices selected for the striatal binding. The specific striatal binding ratio was calculated as follows: Striatal binding ratio= Striatal ROI-Occipital ROI/Occipital ROI.

Statistical analysis

Comparison between quantitative variables was~~ere~~ done using Student t test for independent or paired samples, ~~-~~or Mann-Whitney's test, ~~, and the~~ X^2 or Fisher exact test was used to analyze categorical variables. Multivariate analysis was performed with

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multiple lineal and logistic regressions, for quantitative and categorical variables, respectively. In addition to age and sex, ~~V~~variables with significant differences among groups were included in the models in order to adjust for them. The cutoff value of the ^{123}I-ioflupane SPECT distinguishing PD converters from non-converters was established using a~~n~~ ROC curve. For the evaluation of the olfactory function, mean raw UPSIT scores were compared between groups and afterward the individual scores were categorized (normosmic *vs* hyposmic) using normative data for age and sex as previously reported by Doty[5], with a dichotomous cut at the 15th percentile. Analyses were performed using SPSS 15.0 for Windows (SPSS, Inc., Chicago, IL).

Standard protocol approvals, registrations, and patient consents

All participants gave their informed consent to participate in the study, which was approved by the ethics board of the University Hospital Marqués de Valdecilla–IDIVAL.

Results

From the initial cohort of 32 participants at baseline, three declined to participate in any of the 4-year evaluations and another four were available only for the clinical examination but not for olfaction testing and imaging~~Twenty-nine out of the 32 asymptomatic carriers from the baseline study participated in the 4-year follow-up~~. Mean age of the 29 participants was 64.4 ± 11.9 years, 60.6% female. Mean interval between the baseline and the 4-year clinical and neuroimaging examination was 49.3 ± 4.9 months. Demographic and clinical data of the participants are summarized in table 1~~.~~.

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Clinical evaluation

Three *LRRK2* G2019S mutation carriers, asymptomatic at baseline, had converted to PD at the 4-year evaluation. Cases 10 and 17, aged 58 and 75 years, had been diagnosed of PD in 2015 and 2012, respectively, and case 20, a female aged 91, had been diagnosed of PD in 2013 (in the second year of follow-up). Their UPDRS-III score at the 4-year evaluation were 21.5, 21 and 35, respectively. Two participants ~~subjects~~ (cases 5 and 24) showed minimal signs of PD with a UPDRS-III score of 6 and 4 respectively but didn't fulfill clinical diagnostic criteria of PD. Compared to the mean UPDRS-III score at baseline there was a significant increase in the score at four years in the entire cohort (1.8 ± 4.4 *vs* 4.3 ± 8.2; p= 0.03 ~~OK Student's T for paired samples~~). Mean UPDRS-III in PD-converters was higher, although not significantly, than in non-converters at baseline (9.3 ± 10.7 *vs* 0.8 ± 1.8; p= 0.3 ~~OK Student's T INDEPENDENT SAMPLES VARIANZAS DISTINTAS~~) but was significantly higher at the four-year evaluation (25.8 ±7.9 *vs* 1.6 ± 1.7; p= 0.03 ~~OK Student's T INDEPENDENT SAMPLES VARIANZAS DISTINTAS~~). PD-converters were older than non-converters (74.7 ±16.5 *vs* 63.3 ± 10.6 years ~~; p= 0.10 ok Student's T INDEPENDENT SAMPLES~~), however differences were not statistically significant (p= 0.17).

Olfaction Testing

Twenty-five carriers of the mutation completed the UPSIT test at the end of the follow-up. Mean UPSIT score at 4-years was not significantly different from baseline (28.3 ± 4.4 *vs* 31.1 ± 3.6, p= 0.43). Mean UPSIT decline of the entire cohort during the follow-up was 2.7 points. Mean UPSIT score at baseline was not different between PD-converters and non-converters (29.7 ± 6.6 *vs* 30.9 ± 3.3, p=0.8) and that was also the case at 4-year after adjusting for age (21.3 ± 4.5 *vs* 29.2 ± 3.5; p= 0.02; p-adjusted=

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0.23). Among PD-converters, cases 10 and 17 had a significant decline in their UPSIT score of 17 and 12 points respectively, however case 20 improved her score in 4 points. The proportion of hyposmic carriers at the 4-year evaluation was 44%, higher compared with 28% at baseline, however only one out of the three PD-converters was hyposmic at the end of the 4-year of follow-up.

Dopamine transporter imaging

At the four-year assessment there was a mean absolute reduction of ^{123}I-ioflupane binding ratio in bilateral striatum of 22.6 ± 10.8 % (range: 3.7-47%) compared to baseline measures (1.18 ± 0.35 *vs* 1.51 ± 0.39; p= 1.56 x10^{-10} ~~OK Student's T for paired samples~~). The absolute rate of decline in PD-converters was significantly higher than that observed in non-converters (40.7 ± 5.8% *vs* 19.7 ± 8.8%; p= 0.001 ~~OK Student's T T INDEPENDENT SAMPLES~~); however, the slope of the decline was similar between both groups given that PD-converters has significantly lower ^{123}I-Ioflupane~~FP-CIT~~ uptake at baseline than non-converters (0.91 ± 0.08 *vs* 1.59 ± 0.33, p= 0.002~~Student's T INDEPENDENT SAMPLES~~), ~~(~~Figure 1~~FIGURA 1?~~). At the 4-year scan mean ^{123}I-ioflupane binding ratio was also significantly lower in PD-converters compared to non-converters (0.5~~42~~ ± 0.03 *vs* 1.27~~8~~± 0.27~~8~~,~~)~~ p= 1.83 x10^{-11})~~0.006~~. ~~NO SE DE DONDE SALEN ESOS VALORES PERO CREO QUE HAY UN ERROR Y si te fijas en la tabla de resultados EN VEZ DE CALCULAR ESTO CALCULAMOS SOLO EL LADO IZQUIERDO ("Estriado izdo DTS2")~~ A baseline scan with a ratio of bilateral striatal uptake below 1 predicted with 100% sensitivity and specificity conversion to PD within the four-year period (Area Under the Curve= 1, p~~-~~= 0.006). The mean reduction in the ratio of mean bilateral striatum ^{123}I-ioflupane uptake was 0.33 ± 1.6, with no differences between PD-converters and non-converters (0.37 ± 0.08 *vs* 0.32 ± 0.16, p=

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0.64~~OK Student's T INDEPENDENT SAMPLES~~; p-adjusted [age, sex, time interval between ^{123}I-ioflupane SPECTs]= -0.48~~2~~). ~~lineal regression ese p-value no se muy bien de donde sale, he vuelto a hacer la regresion y sería 0.48?.~~ The variable time-adjusted decline in ^{123}I-ioflupane uptake observed between scans was not correlated with any of the variables studied (age, sex, baseline SN echogenicity or UPSIT score). There was a significant correlation between mean bilateral striatal ^{123}I-ioflupane uptake and UPDRS-III at 4-years (r= -0.65, p=0.001~~OK~~, p-adjusted= 0.006)~~ OK~~. Mean bilateral striatal ^{123}I-ioflupane uptake at 4-years was also correlated with age (r= -0.42, p=0.03). On the contrary, no correlation was observed between the mean maximum area of SN echogenicity and mean bilateral striatal ^{123}I-ioflupane uptake at baseline (r= -0.20, p=0.33).

Discussion

In this study we provide information supported on prospective clinical and imaging data on the premotor stage of *LRRK2* G2019S-associated PD. After 4 years of follow-up 12% of carriers from a cohort aged 59 years at baseline, converted to PD. The ratio of mean bilateral striatum dopamine transporter (DaT) binding below 1 on baseline ^{123}I-ioflupane SPECT predicted with a high sensitivity and specificity conversion to motor PD within the 4-year period. The slope of DaT binding decline over the 4-year period was not different between PD-converters and non-converters, this implying that most likely the rate of decline is constant along the premotor and earliest clinical stages of the disease.

The neurodegenerative process of PD patients carrying the *LRRK2* G2019S mutation results in a pattern of nigrostriatal dopaminergic dysfunction similar to that observed in ~~i~~idiopathic PD, with identical neurochemical phenotype reported on

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dopamine transporter imaging[6]. Although progressive decline in striatal dopamine transporter binding in PD is not a direct measure of changes in SN cell counts, results in previous studies are in accordance with postmortem data[7]

Previous studies have shown a linear reduction of striatal DaT binding in early PD, with estimated annual rates of decline ranging from 4% to 10%[8-12]. In our study, assuming a constant decline, the estimated absolute annual rate of decline of striatal dopamine transporter binding was around 5%. It is important to highlight however that~~In~~ we couldn't calculate the relative rate of decline compared to age-expected normal values at the time of the first scan, since we didn't have that data. Considering this, the relative annual DaT binding decline in our study would be below 4%, slightly lower than previous studies performed in early idiopathic PD. The slope of the decline was quite similar between carriers that converted to PD within the 4-year period and those who didn't; however, the slope of the decline of striatal DaT binding was widely variable, with some participants ~~subjects~~ showing softer slopes of decline than others (see Fig 1). This variability was not correlated with any of the variables studied here. Indeed, participants~~subjects~~ with a milder slope of decline of DaT binding were no different in terms of age, gender or DaT binding at baseline compared to those with a more pronounced slope of decline~~. Even among sibling pairs differences in terms of baseline striatal dopamine transporter uptake and slope of the decline was observed, this highlighting the existence of still unknown modifiers of disease penetrance and progression~~. Our data also indicate that the initiation of dopaminergic terminal loss probably antedates more than four years the onset of motor symptoms, since 75% of carriers with visually abnormal DaTSCAN at baseline didn't develop motor symptoms of PD along the 4-year period. A recent study based on serial dopamine transporter imaging has estimated the duration of the pre-symptomatic stage of PD in

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approximately 10 years[13].

Very few studies have reported longitudinal data on DaT imaging in asymptomatic carriers of *LRRK2* mutations. One study reported~~Adams JR, et al reported~~ on the case of two asymptomatic carriers (aged < 55 years) of *LRRK*2 mutations (different from G2019S) showing a decline of 14% in striatal DaT binding in two serial PET scans separated four years[14]. In another study, two asymptomatic carriers of the R1441C mutation showed no decline on 11C-MP-PET binding in two studies separated 21-36 months[15].

Several cross-sectional studies have already provided information re~~a~~garding olfaction as a potential biomarker in *LRRK2* G2019S-associated PD. On the one hand hyposmia has been shown to be approximately 30% less frequent in *LRRK2*-PD than in idiopathic PD[3,16]. On the other hand, asymptomatic carriers of the *LRRK2* G2019S mutation are not more hyposmic than their relatives non-carriers and healthy controls[3,17]. Our data, coming from a longitudinal study, also suggest that olfaction loss is not common at the earliest stages of *LRRK2* G2019S-associated PD and therefore is not a good predictor of phenoconversion to PD at the premotor stage. Indeed, two out of the three PD-converters from our study were not hyposmic by the time they developed PD motor symptoms. Interestingly however, two of PD-converters showed a significant decline in their UPSIT score between the baseline and the 4-year evaluation. Interpreted in the context of the established notion that around 50% of *LRRK2*-PD patients from different cohorts are hyposmic, this suggests that hyposmia in such~~this~~ cases most likely develops shortly before motor symptoms appear or soon after this moment.

We had previously shown that an increased area of SN echogenicity was a constant feature (present in 90% of cases) in *LRRK2* G2019S asymptomatic carriers[3]. As it was anticipated this feature was not predictive of phenoconversion to PD within

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the 4-year period in this study. We also didn't find a correlation between the maximum area of SN echogenicity and either the baseline striatal DaT uptake or the slope of its decline over time. This is in accordance with data coming from different studies that indicate that SN echogenicity is not a marker of disease progression but rather a stable feature probably indicating vulnerability of the nigrostriatal system[18].

Age is the only variable known to correlate with disease penetrance in carriers of the G2019S mutation of the *LRRK2* gene. Penetrance increases with age until 25-75% at 80 years according to different studies[2,19,20]. As expected, PD-converters in our study were older (an average of ten years) than non-converters, with statistical non-significance probably just because of the low number of cases. One of the cases however was below 60 years when he developed PD motor symptoms.

Our study has several limitations. The main limitation is that the sample is not large and as a consequence only three carriers converted to PD despite the long follow-up. This might affect the reproducibility of our findings in other cohorts of *LRRK2* carriers. Also, we cannot completely exclude that dopaminergic drugs had influenced the results of ^{123}I-ioflupane SPECT in PD-converters, since they were already on PD drugs when the second scan was performed. We acknowledge that some studies have suggested that L-dopa might down-regulate striatal DaT binding, however we consider unlikely this had biased our results given that two out of the three PD-converters were not taking L-dopa (on low doses of dopamine agonists) and the three of them showed similar slopes of decline, also comparable to most of non-converters. Also, as acknowledged before, estimation of the rate of decline of DaT binding over time would have been better interpreted in relation relative to data of age - expected normal values, which we didn't have. We also acknowledge that a third point of evaluation and imaging acquisition will be necessary to demonstrate whether the rate of decline of DaT

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binding along time is constant, as we suspect based on our data, or not. For this purpose we plan to do a further evaluation of this cohort in a three-year period. This planned follow-up will be also necessary to confirm whether the proposed cutoff value of < 1 on the ratio of mean bilateral striatal DaT binding can be used as reliable predictor of early conversion to PD in this cohort.

We consider that the results of this study might be of some help for different aspects related with the design of disease modifying drug trials and also for genetic counseling~~.~~ ~~Clarifying what's the duration of the prodromal stage of PD and determining the rate of decline of dopaminergic terminals is of crucial importance for planning clinical trials with drugs aiming to modify disease progression~~. It is widely accepted that the earliest stages of the disease would be the ideal scenario for testing these drugs, before much of the damage is already done and therefore with higher chances of success. Asymptomatic carriers of the *LRRK2* G2019S mutation are considered an ideal population ~~target population~~ for ~~developing these drugs~~ this purpose ~~just because they constitute a very homogeneous population and also because they~~ since patients could be recruited in an early premotor stage of the disease. ~~Ideally, results from these eventual trials would be extrapolated to the most common forms of idiopathic PD, given their great similarities.~~ There are some issues however; it's a fact that although *LRRK2* G2019S carriers are in a high risk of developing PD, nearly 50% escape the disease. We therefore need tools to reliably predict which carriers will develop motor symptoms during life and which among them are closest in time to PD-conversion. Although findings from this study, due to small sample size, cannot be conclusive and might be non-reproducible, ~~Here~~ we have shown that in addition to age the ratio of striatum dopamine transporter binding on ^{123}I-ioflupane SPECT could be a useful tool for that purpose. According to this notion, asymptomatic carriers with striatal DaT uptake below an established cutoff value might

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be the best candidates for participating in these clinical trials. Also, if confirmed in further follow-up evaluations of this cohort and others this information would be of great importance for counseling more accurately carriers about their risk of developing Parkinson's disease. The imaging data from this paper highlight the need to further explore dopamine transporter imaging with ^{123}I - ioflupane as a screening tool for those at risk of PD and as a biomarker for monitoring disease progression.

~~**Author contributions**~~

~~Dr. Sierra: drafting/revising the manuscript, study concept or design, analysis or interpretation of data, acquisition of data. Dr. S~~[illegible]~~chez-Juan: drafting/revising the manuscript, analysis or interpretation of data, statistical analysis. Dr. Mart~~[illegible]~~ez-Rodr~~[illegible]~~uez: drafting/revising the manuscript, analysis or interpretation of data, acquisition of data. Dr. Gonz~~[illegible]~~lez-Aramburu: drafting/revising the manuscript, acquisition of data. Dr. Jim~~[illegible]~~ez-Alonso: drafting/revising the manuscript, analysis or interpretation of data, acquisition of data. Dr. S~~[illegible]~~nchez-Rodr~~[illegible]~~uez: drafting/revising the manuscript, acquisition of data. Dr. Banzo: drafting/revising the manuscript, analysis or interpretation of data, acquisition of data. Dr. Infante: drafting/revising the manuscript, study concept or design, analysis or interpretation of data, acquisition of data, study supervision, obtaining funding.~~

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~~report no disclosures.~~

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Table 1

Clinical and dopamine transporter imaging, baseline and 4-year, in asymptomatic carriers of *LRRK2* G2019S mutation: PD converters *vs* non-converters.

	Asymptomatic carriers (n=22)	PD-converters (n=3)	P

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Age, years (SD)	63 (9.7)	74.3 (16)	0.17
Sex (%females)	64	33.3	0.34
UPDRS-III, Baseline, mean (SD)	0.8 (1.8)	9.3 (10.7)	0.08
UPDRS-III, 4-year, mean (SD)	1.6 (1.7)	25.8 (7.9)	0.004
UPSIT score, Baseline, mean (SD)	30.8 (3.3)	29.7 (6.7)	0.85
UPSIT score, 4-year, mean (SD)	29.1 (3.6)	21.3 (4.5)	0.02 0.23
UPSIT decline, Baseline to 4-year, mean (SD)	2.3 (3.8)	6.5 (14.8)	0.91
Striatal DaT binding ratio, Baseline, mean (SD)*	1.6 (0.3)	0.91 (0.1)	0.006
Striatal DaT binding ratio, 4-year, mean (SD)*	1.26 (0.27)	0.5 (0.03)	0.006
DaT binding decline, Baseline to 4-year, mean (SD)	0.33 (0.16)	0.37 (0.08)	0.65
Maximum area of SN echogenicity (cm^2), mean (SD)	0.27 (0.07)	0.27 (0.03)	0.80
Abnormal ^{123}I-ioflupane SPECT, Baseline, %	40	100	0.09
Abnormal ^{123}I-ioflupane SPECT, 4-year, %	70.8	100	0.34
Hyposmic participants, Baseline, %	32	0	0.04
Hyposmic participants, 4-year, %	43	33.3	0.62

* Striatal ROI-Occipital ROI/Occipital ROI

** P-adjusted for age and sex

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~~Table 2~~

~~**Clinical and dopamine transporter imaging, baseline and 4-year, in asymptomatic carriers of *LRRK2* G2019S mutation: PD converters vs non-converters.**~~

	~~Asymptomatic carriers (n=22)~~	~~PD-converters (n=3)~~	~~P~~
~~Age, years (SD)~~	~~63 (9.7)~~	~~74.3 (16)~~	~~0.17~~
~~Sex (M/F) (mejor poner %females no?)~~	~~36/64~~	~~66.7/33.3~~	~~0.34~~
~~UPDRS-III, Baseline, mean (SD)~~	~~0.8 (1.8)~~	~~9.3 (10.7)~~	~~0.08~~
~~UPDRS-III, 4-year, mean (SD)~~	~~1.6 (1.7)~~	~~25.8 (7.9)~~	~~0.004~~
~~UPSIT score, Baseline, mean (SD)~~	~~30.8 (3.3)~~	~~29.7 (6.7)~~	~~0.85~~
~~UPSIT score, 4-year, mean (SD)~~	~~29.1 (3.6)~~	~~21.3 (4.5)~~	~~0.02~~
~~UPSIT decline, Baseline to 4-year, mean (SD)~~	~~2.3 (3.8)~~	~~6.5 (14.8)~~	~~0.91~~
~~Striatal DaT binding ratio, Baseline, mean (SD)*~~	~~1.6 (0.3)~~	~~0.91 (0.1)~~	~~0.006~~

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Striatal DaT binding ratio, 4-year, mean (SD)*	1.26 (0.27)	0.5 (0.03)	0.006
DaT binding decline, Baseline to 4-year, mean (SD)	0.33 (0.16)	0.37 (0.08)	0.65
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Abnormal ^{123}I-ioflupane SPECT, Baseline, %	40	100	0.09
Abnormal ^{123}I-ioflupane SPECT, 4-year, %	70.8	100	0.34
Hyposmic subjects, Baseline, %	32	0	0.34
Hyposmic subjects, 4-year, %	43	33.3	0.62

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* Striatal ROI-Occipital ROI/Occipital ROI

** Adjusted for age and sex la forma directa de ajustar estas p es por regresion logistica, pero ya sabes que nos era imposible realizarla dada la baja N de convertidores. mejor dejarlo tal cual.

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~~**Table 1**~~

~~**Cohort of *LRRK2* G2019S carriers: clinical, demographic and dopamine transporter imaging data, baseline and 4-years follow-up**~~

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Case	Sex	Age	SN echo (cm²)	Altered DaTSCAN (Baseline)	Altered DaTSCAN (4 year)	Mean Striatal DaT binding (Baseline)	Mean Striatal DaT binding (4 year)	UPSIT Baseline	Hyposmia Baseline
1	M	71	0.30	Yes	–	–	–	32	No
2	F	46	0.14	Yes	Yes	1.64	1.17	36	No
3	M	86	0.38	Yes	–	–	–	6	Yes
4	F	53	0.27	No	–	–	–	30	Yes
5	F	74	0.30	No	No	1.44	1.02	30	No
6	F	66	0.29	No	Yes	1.81	1.26	32	No
7	F	59	0.31	No	No	1.92	1.74	32	Yes
8	F	57	0.36	No	No	1.75	1.64	34	No
9	F	67	0.26	Yes	Yes	1.82	1.41	32	No
10	M	58	0.30	Yes	Yes	0.82	0.53	34	No
11	M	66	0.31	No	Yes	1.46	1.01	32	No
12	M	55	0.34	No	No	1.58	1.46	29	Yes
13	M	88	0.26	Yes	–	–	–	25	No
14	F	60	0.18	Yes	Yes	1.88	1.50	28	Yes
15	F	68	0.27	Yes	Yes	1.09	1.05	26	Yes
16	F	69	0.11	No	Yes	1.52	1.14	30	Yes
17	M	75	0.25	Yes	Yes	0.95	0.57	33	No
18	M	56	0.25	Yes	Yes	1.18	0.94	33	No
19	F	53	0.21	No	–	–	–	33	No
20	F	91	–	Yes	Yes	0.96	0.51	22	No
21	F	61	0.3	No	Yes	1.04	0.81	36	No
22	F	53	0.18	No	Yes	1.98	1.43	34	No
23	M	75	0.38	Yes	Yes	1.05	0.92	29	No
24	F	67	0.26	No	No	2.31	1.76	30	Yes
25	M	81	0.25	Yes	–	–	–	23	No
26	F	55	0.26	Yes	Yes	1.82	1.28	35	No
27	M	50	0.20	No	–	–	–	28	Yes
28	F	72	0.22	No	Yes	1.77	1.44	31	No

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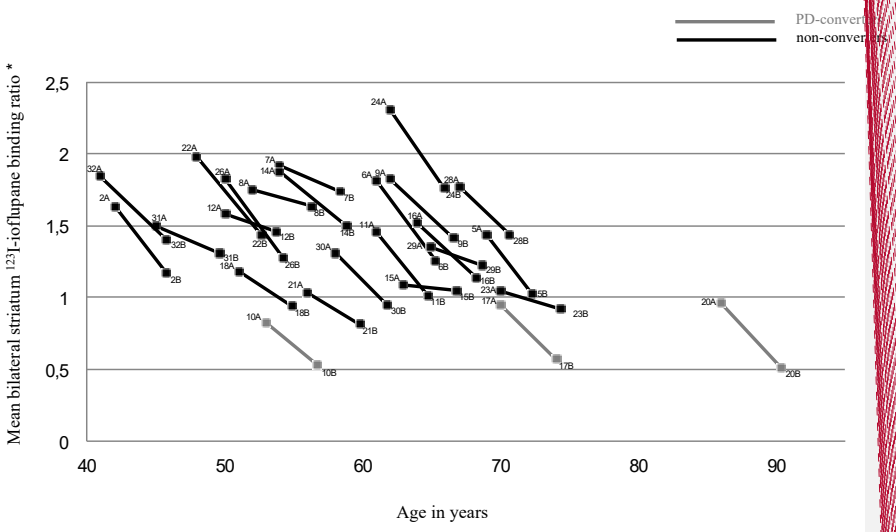
29	F	71	0.33	No	Yes	1.35	1.22	29	No	2
30	M	63	0.36	No	Yes	1.31	0.95	30	No	2
31	F	49	0.23	No	No	1.50	1.31	34	No	2
32	F	46	0.40	No	No	1.85	1.40	33	Yes	3

Figure 1

Individual decline of mean bilateral striatum ^{123}I-ioflupane binding ratio between baseline and 4-year



* Striatum count number-occipital count number/occipital count number

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