The association between clinical phenotype of Parkinson's disease and LRRK2 variants in China

shishuang cui
Shanghai Jiao Tong University Medical School Affiliated Ruijin Hospital
gen li
Shanghai Jiao Tong University Medical School Affiliated Ruijin Hospital
yiqi lin
Shanghai Jiao Tong University Medical School Affiliated Ruijin Hospital
yixi he
Shanghai Jiao Tong University Medical School Affiliated Ruijin Hospital
pingchen zhang
Shanghai Jiao Tong University Medical School Affiliated Ruijin Hospital
guiying he
Shanghai Jiao Tong University Medical School Affiliated Ruijin Hospital
qian sun
Shanghai Jiao Tong University Medical School Affiliated Ruijin Hospital
yuyan tan
Shanghai Jiao Tong University Medical School Affiliated Ruijin Hospital
shengdi chen (chen_sd@medmail.com.cn)
Shanghai Jiao Tong University Medical School Affiliated Ruijin Hospital

Research article

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Abstract

Introduction: LRRK2 G2385R and LRRK2 S1647T have been identified as the most common risk variants for PD in the Chinese population. The aim of the study was to explore the correlation of LRRK2 G2385R, LRRK2 S1647T and their haplotypes with symptoms.

Method: Demographic variables, disease-related variables and motor and non-motor assessments was collected in the study. Peripheral blood samples were collected, and DNA was extracted. SNaPShot technique was used to analysis DNA genotype. Chi square test and ANOVA was used to test the between-group differences. Risk analysis was performed by logistic regression model or Cochran-Armitage model.

Results: 502 PD patients were enrolled in the study. The scores of PDSS and MoCA were significant higher in LRRK2 S1647T variants carriers genotype after adjustment. The scores of BPI, attention in NMSS, cardiovascular in SCOPA-AUT was significant lower in LRRK2 G2385R variants carriers adjusted for H-Y stage and gender. LRRK2 ARG 2385 was associated with reduced risk of sialorrhea (p=0.049, additive model) and postural hypotension (p=0.030, additive model; OR=0.35, 95%CI: 0.10-0.89, adjusted P=0.050, dominant model). rs11564148A rs34778348A was also found associated with a reduced risk with postural hypotension adjusted for H-Y staging and gender.

Conclusion: Our study indicated that LRRK2 S1647T variants carriers presented better motor symptoms, sleep quality and cognition. LRRK2 G2385R variants carriers presented better autonomic function and cognition and had a reduced risk of sialorrhea and postural hypotension. rs11564148A - rs34778348A was associated with reduced risk with postural hypotension.

1. Backgrounds

Parkinson's disease (PD) is the second common neurodegenerative disease. The average prevalence of PD in China was about 3.8756‰ (≥50 years) in Han population, bringing a huge burden to Chinese economics and healthcare system[1]. The clinical presentation and course of Parkinson's disease (PD) is heterogeneous, with variability in onset, progression, motor and non-motor symptoms. Mutations in the leucine-rich repeat kinase 2 (*LRRK2)* gene are the most frequent genetic cause associated with autosomal dominant PD, accounting for about 14% of PD in the Chinese individuals[2]. Although the *LRRK2* G2019S variants is the most common in several ethnic populations worldwide, this variant is very rare in Asian populations. *LRRK2* G2385R and *LRRK2* S1647T have been identified as the most common risk variants for PD in the Asian population[3]. Thus, whether these variants confer a different disease phenotype need to be explored.

Previous studies reported no significant correlation of the *LRRK2* G2385R variant with motor or non-motor symptoms except for non-significant milder non-motor symptoms in the Chinese population[2]. Rare studies explored the correlation of the LRRK2 S1647T variants with symptoms. A longitudinal study found risk variant carriers of *LRRK2* G2385R, R1628P and S1647T experienced greater rate of motor progression than noncarriers[4]. As the result of linkage disequilibrium, haplotype of *LRRK2* was much

less mentioned. A Taiwanese study found that the frequency of 1647T–2385R–2397T haplotype in PD patients was still higher than in control subjects[5]. Previous two studies both found that all patients who were G2385R and/or R1628P carrier also carried the S1647T variant [4, 6]. A study indicated that age at onset of variants of *LRRK2* R1628P + S1647T or G2385R + S1647T was not significant different from noncarriers[6]. But the clinical phenotype of haplotype of *LRRK2* 1647T–2385R has not been discussed separately. In our previous study, we found the *LRRK2* G2385R variant could be a risk factor for the PIGD phenotype, motor fluctuations, LED values and RBD symptoms in PD patients. But the features of *LRRK2* S1647T variant and the *LRRK2* haplotypes was not explored in the previous study[7]. Thus, in this study, we aimed to explore the clinical features of PD patients with the *LRRK2* S1647T variant and the *LRRK2* haplotypes. Besides, we also explored the clinical features of PD patients with the *LRRK2* G2385R variant with a larger sample.

2. Methods

2.1 Participants

The participants in our study were enrolled between 2016 and 2018 from Movement Disorders Clinic at the Department of Neurology, Rui Jin Hospital affiliated to Shanghai Jiao Tong University School of Medicine. All patients were diagnosed with PD by movement disorders specialists, according to the criteria of Movement Disorder of Society[8]. Exclusion criteria included secondary parkinsonism, atypical parkinsonism and other movement disorders other than PD. Commorbities that might interfere with the reliable completion of clinical assessments such as severe hearing or visual loss, inability to speak or write were also excluded. Participants were fully informed and signed consent form before the study. The study was approved by the medical ethics committee of Rui Jin Hospital affiliated to Shanghai Jiao Tong University School of Medicine.

2.2 Assessments

Demographic variables including age, sex and schooling year were recorded during a clinical interview. Disease-related variables including age at onset (AAO), disease duration and drugs were collected. Disease stage was assessed with the Hoehn &Yahr staging (H-Y stage). Family history of PD was also collected. Disease-related decline in non-motor function, activity of daily living (ADL) and motor function were assessed with the Movement Disorder Society Unified Parkinson's Disease Rating Scale (MDS-UPDRS) part 1, part 2 and 3. Life quality was assessed by the 39-item Parkinson disease questionnaire (PDQ–39).

Non-motor symptoms were assessed by Non-motor Symptoms Scale (NMSS). Cognitive function was assessed with the Mini-mental State Examination (MMSE) and Montreal Cognitive Assessment (MoCA) Beijing Version. Depression and anxiety were quantified with the 17-item Hamilton Depression Rating Scale (HAMD) and Hamilton Anxiety Rating Scale (HARS), respectively. Olfactory function was assessed with 16-item odor identification test from the extended version of sniffin' sticks (SS–16) and hyposmia was considered when SS–16 < 8.3[9]. Autonomic function was assessed with the scale for outcomes in

PD for autonomic symptoms (SCOPA-AUT). Rapid eye movement sleep behavior disorder (RBD), and excessive daytime sleepiness (EDS) were assessed with the RBD Questionnaire-Hong Kong (RBD-HK) and Epworth Sleepiness Scale (ESS), separately. Probable RBD was defined that the score of RBD-HK was more than 17[10]. Sleep quality was assessed by Parkinson's disease sleep scale (PDSS). Pain and fatigue were assessed by the brief pain inventory (BPI) and fatigue severity rating scale (FSS).

2.3 Genetic analysis

Peripheral blood samples were collected, and DNA was extracted from leukocytes using phenol–chloroform method[11]. The Primer Premier 5 (version 5.00, PREMIER Biosoft International) was used to design primers for *LRRK2* G2385R and *LRRK2* S1647T. Phosphorylase (FastAP) and exonuclease I (EXO I) were adopted to purify the polymerase chain reaction (PCR) products of these 2 SNPs. ABI SNaPshot Multiplex kit was then used to extend them and further purified by FastAP, loaded on ABI3730xl subsequently. GeneMapper 4.0 (version 4.0, Applied Biosystems) was used to analysis the SNP genotypes.

2.4 Statistical analysis

Statistical analyses were performed with SPSS Statistics (version 20.0, SPSS Inc., Chicago, IL, USA). Continuous variables were given as means and standard deviation. Categorical variables were summarized by percentages. Chi square test was performed to test distribution differences of gender, education level, H-Y stage, family history among PD patients with *LRRK2* variants. Analysis of Variance (ANOVA) was used to test the difference of age, scores of different scales adjusted for H-Y stage and gender except MMSE and MoCA adjusted for gender, H-Y stage and education level. Risk analysis was performed by logistic regression model or Cochran-Armitage model. Cochran-Armitage was used in additive model adjusted for gender and H-Y stage. Logistic regression was used for dominant, recessive and overdominant models adjusted for gender and H-Y stage. Odds Ratio (OR), 95% Confidence Interval (CI), and p-value (two-tailed test) were computed. Significance of differences was defined as two-tailed $p < 0.05$.

3. Results

502 PD patients were enrolled in the study, among which 61 PD patients had family history. No significant differences of age, gender, family history and education level were found among PD patients with different *LRRK2* S1647T genotypes and among PD patients with different *LRRK2* G2385R genotypes (supplementary table 1). 54 patients were both G2385R and R1628P carriers. No significant differences of hypertension, diabetes, coronary heart disease, smoking and drinking were found among PD patients with different *LRRK2* S1647T genotypes and among PD patients with different *LRRK2* G2385R genotypes (supplementary table 2).

Among *LRRK2* S1647T genotypes, the scores of PDSS and MoCA were significant higher in variants carriers after adjustment (table 1). Among the *LRRK2* G2385R genotypes, the scores of BPI, attention in

NMSS, cardiovascular in SCOPA-AUT was significant lower in variants carriers adjusted for H-Y stage and gender (table 2). The scores of MMSE was significant higher in variants carriers adjusted for H-Y stage, gender and disease duration (table 2).

A further risk analysis revealed that no symptoms were associated with *LRRK2* Thr^{1648} after adjustment (supplementary table 3). *LRRK2* Arg^{2385} was associated with reduced risk of sialorrhea (p = 0.049, additive model) and postural hypotension (p = 0.030, additive model; OR = 0.35, 95% CI: 0.10 - 0.89, adjusted p = 0.050, dominant model) (table 3).

Haplotype block of rs11564148A - rs34778348A was also found associated with a reduced risk with postural hypotension adjusted for H-Y staging and gender compared with haplotype block of rs11564148 T - rs34778348 G (OR = 0.27, 95%CI: 0.06–0.79, p = 0.035, table 4). We did not perform the analysis between haplotype block of rs11564148 T - rs34778348 A and reference haplotype due to low sample amount of haplotype block of rs11564148 T - rs34778348 A (n = 2).

4. Discussion

In this study, we found that *LRRK2* S1647T variants carriers were associated with better motor symptoms, sleep quality and cognition. Variants carriers of *LRRK2* G2385R were associated with better autonomic function and cognition and less pain. *LRRK2* Arg^{2385} was associated with a reduced risk of sialorrhea and postural hypotension. Haplotype block of rs11564148 A - rs34778348 A was associated with a reduced risk of postural hypotension.

The *LRRK2* G2835R variant is a common polymorphism and is associated with a two-fold increased risk of PD in Singaporean and Taiwan Chinese populations [12, 13]. A study of a Chinese cohort in mainland found 13.1% carried LRRK2 G2385R and a 1.65-fold increase risk of PD[14]. Several studies explored the association between *LRRK2* G2385R and symptoms in PD and found no significant differences in the motor and non-motor symptoms[2, 15, 16].Consistent with our previous study, the MMSE score was also higher in the G2385R variant carrier group. Similar with the previous study, no significant differences were found in SCOPA-AUT scores. Furthermore, we analyzed the association between subscores of SCOPA-AUT and *LRRK2* G2385R and found the scores of cardiovascular in SCOPA-AUT was significant different in our study. A further risk analysis in our study was performed and found that *LRRK2* G2385R variants carriers was associated with a reduced risk of postural hypotension.

The *LRRK2* S1647T polymorphism is found common in Chinese population. A previous study found that the homozygous S1647T genotype (AA) was associated with a 1.815-fold increased risk of PD in Southern China and *LRRK2* variant S1647T was identified as a risk factor for PD development in a Taiwanese study [17, 18]. But its influence on the clinical features of PD still remains to be elucidated. In our study, AA genotype of *LRRK2* S1647T were associated with higher scores of MoCA but not MMSE. Consistent with the study by Zheng and his colleagues, no significant differences in MMSE scores between carriers and non-carriers[19]. They also performed Stroop word color test (SWCT) to evaluate

executive function and found that the SWCT-TIME scores of LRRK2 S1647T carriers were significantly lower than those of LRRK2 S1647T noncarriers[19]. As MoCA have more detailed assessment in executive function compared with MMSE, the founding also supported the result in our study. In our study, LRRK2 S1647T carriers had a better sleep quality according to the PDSS scores. None have studied the association between sleep and LRRK2 S1647T before. Only one study studied RBD and LRRK2 between RBD patients and control and found that LRRK2 S1647T,was associated with risk for RBD but the association disappear after correction for multiple comparison[20].

To our knowledge, this is first study to analysis the association between symptoms and haplotypes LRRK2. We found that rs11564148A - rs34778348A was associated with a reduced risk with postural hypotension. The scores of cardiovascular in SCOPA-AUT was lower in LRRK2 S1648T variants carriers in trend and significantly lower in LRRK2 G2385R variants carriers in our study. Thus, they may explain the result in our study.

Limitations should be considered in interpreting our findings. The sample in our study was relatively small and thus a selection bias should be considered. Secondly, we only detect two loci of *LRRK2* in PD patients, and thus non-carrier might include individual with other locus of *LRRK2* gene and other genetic variants. Effects of other genetic variants cannot be excluded in our study. Furthermore, we only explored the association between LRRK2 variants and symptoms rather than the severity of symptoms. In addition, the assessment of symptoms was not objective. Thus, further study is needed to enlarge the sample size and assess the severity of symptoms.

5. Conclusion

In summary, our study indicated that *LRRK2* S1647T variants carriers may presented better motor symptoms, sleep quality and cognition. *LRRK2* G2385R variants carriers may presented better autonomic function and cognition and had a reduced risk of sialorrhea and postural hypotension. Haplotype block of rs11564148A - rs34778348A was associated with reduced risk with postural hypotension. But further studies with larger sample and more detailed assessment of the severity of symptoms are needed.

Abbreviations

AAO, age at onset

ADL, activity of daily living

ANOVA, Analysis of Variance

BPI, brief pain inventory

CI, confidence interval

EDS, excessive daytime sleepiness

ESS, Epworth Sleepiness Scale

EXO I, exonuclease I

FSS, fatigue severity rating scale

HAMD, 17-item Hamilton Depression Rating Scale

HARS, Hamilton Anxiety Rating Scale

H-Y stage, Hoehn &Yahr staging

LRRK2, leucine-rich repeat kinase 2

MDS, movement disorder society

MMSE, Mini-mental State Examination

MoCA, Montreal Cognitive Assessment

NMSS, Non-motor Symptoms Scale

OR, odds ratio

PCR, polymerase chain reaction

PD, Parkinson's disease

PDQ–39, 39-item Parkinson disease questionnaire

PDSS, Parkinson's disease sleep scale

RBD, Rapid eye movement sleep behavior disorder

RBD-HK, Rapid Eye Movement sleep behavior disorder questionnaire-Hong Kong

SCOPA-AUT, scale for outcomes in Parkinson's disease for autonomic symptoms

SCWT, Stroop word color test

SD, Standard deviation

SS–16, 16-item odor identification test

UPDRS, Unified Parkinson's Disease Rating Scale

Declarations

Ethics approval and consent to participate

Participants were fully informed and signed consent form before the study. The study was approved by the medical ethics committee of Rui Jin Hospital affiliated to Shanghai Jiao Tong University School of Medicine.

Consent for publication

Not applicable.

Availability of data and materials

The datasets during and/or analysed during the current study available from the corresponding author on reasonable request.

Competing interests

The authors declare that the research was conducted in the absence of any commercial or financial relationships that could be construed as a potential conflict of interest.

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Author contributions

SSC and GL did genetic analysis, performed the statistical analysis and drafted the manuscript. YQL, YXH, PCZ and GYH collected information of Parkinson's disease. SQ did genetic analysis. TYY designed this study and revised the manuscript. SDC designed this study, double-checked the statistical analysis and revised the manuscript.

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Tables

Table 1 the association between rating scales and genotype of rs11564148 of *LRRK2*

	TT genotype (n = 217)	AT genotype (n = 237)	AA genotype (n = 48)	*p* value	*Adjusted p* value
SS-16 (mean ± SD) [a]	5.45 (4.11)	5.26 (4.18)	6.10 (3.94)	0.640	0.484
HAMA (mean ± SD) [a]	5.99 (6.18)	5.58 (5.70)	5.04 (4.92)	0.270	0.390
HAMD (mean ± SD) [a]	5.31 (5.62)	4.86 (4.91)	4.15 (3.95)	0.136	0.284
BPI (mean ± SD) [a]	8.07 (11.91)	9.31 (12.72)	6.47 (8.97)	0.994	0.838
RBD-HK (mean ± SD) [a]	13.05 (16.62)	11.57 (17.16)	11.33 (15.27)	0.346	0.496
PDSS (mean ± SD) [a]	116.00 (23.57)	120.04 (22.07)	123.36 (21.20)	0.026	0.016
PDQ39 (mean ± SD) [a]	14.81 (16.26)	15.31 (17.85)	12.81 (13.96)	0.712	0.971
FSS (mean ± SD) [a]	24.29 (20.03)	22.84 (19.45)	22.17 (18.13)	0.378	0.570
ESS (mean ± SD) [a]	5.84 (6.28)	4.67 (5.63)	6.33 (5.73)	0.481	0.704
MDS-UPDRS (mean ± SD) [a]	45.25 (25.88)	43.47 (26.32)	39.20 (23.29)	0.179	0.109
PART I	8.67 (6.23)	7.56 (5.71)	7.57 (5.33)	0.090	0.112
PART II	10.55 (6.86)	10.15 (7.54)	9.11 (5.78)	0.251	0.129
PART III	26.75 (16.26)	25.48 (17.57)	22.52 (16.81)	0.150	0.093
NMSS (mean ± SD) [a]	29.41 (33.21)	26.19 (32.12)	23.96 (22.99)	0.191	0.300
cardiovascular	0.58 (1.95)	0.59 (2.02)	0.27 (0.71)	0.484	0.821
sleep	6.63 (7.94)	5.20 (6.95)	5.73 (5.95)	0.109	0.168
mood disorder	5.80 (10.85)	5.40 (10.34)	4.52 (7.32)	0.447	0.509
delusion	0.44 (2.73)	0.45 (1.98)	0.21 (0.65)	0.659	0.736
attention	2.37 (3.45)	2.03 (3.30)	2.06 (2.91)	0.326	0.373
gastrointestinal	3.05 (5.11)	2.68 (4.91)	2.42 (3.87)	0.328	0.445
urinary	4.96 (8.59)	3.83 (7.54)	3.69 (5.52)	0.134	0.194
sexual dysfunction	1.31 (4.64)	1.15 (4.25)	0.44 (1.37)	0.264	0.279
others	4.33 (6.07)	4.43 (6.20)	4.63 (6.13)	0.766	0.434
SCOPA-AUT (mean ± SD) [a]	8.98 (9.01)	7.81 (8.74)	7.29 (7.11)	0.110	0.184
gastrointestinal	2.88 (3.40)	2.64 (3.60)	2.50 (3.02)	0.383	0.500
urinary	3.43 (4.44)	2.81 (4.16)	2.73 (3.38)	0.124	0.191
cardiovascular	0.44 (1.22)	0.27 (0.69)	0.19 (0.57)	0.028	0.062
skin	1.64 (2.56)	1.43 (2.38)	1.56 (2.42)	0.544	0.747
sexual dysfunction	0.36 (1.32)	0.29 (1.13)	0.10 (0.52)	0.194	0.214
MMSE (mean ± SD) [b]	26.82 (2.96)	27.03 (3.29)	27.86 (3.00)	0.085	0.061

MoCA (mean ± SD) [b]	22.58 (4.74)	22.98 (4.75)	24.54 (4.03)	0.033	0.016

BPI, brief pain inventory; ESS, Epworth Sleepiness Scale; FSS, Fatigue severity scale; HAMA, Hamilton anxiety rating scale; HAMD, Hamilton depression rating scale; MDS, movement disorders society; MMSE, Mini-Mental State Examination; MoCA, Montreal Cognitive Assessment; NMSS, Non-Motor Symptoms Scale; PDQ-39, 39-item Parkinson's Disease Questionnaire; PDSS, Parkinson's disease sleep scale; RBD-HK, rapid eye movement sleep behavior disorder questionnaire - Hong Kong version; SCOPA-AUT, Scales for Outcomes in Parkinson's Disease-Autonomic questionnaire; SD, standard deviation; SS-16, Sniffin' Sticks 16; UPDRS, Unified Parkinson's Disease Rating Scale
a Hoehn Yahr Staging and gender were taken as adjustment
b Hoehn Yahr Staging, gender and education level were taken as adjustment

Table 2 the association between rating scales and genotype of rs34778348 of *LRRK2*

	GG genotype (n = 445)	mutation carriers (GA + AA) (n = 54 + 2)	*p* value	*p* value
SS-16 (mean ± SD) [a]	5.44 (4.13)	5.27 (4.19)	0.767	0.925
HAMA (mean ± SD) [a]	5.85 (5.98)	4.61 (4.57)	0.136	0.167
HAMD (mean ± SD) [a]	5.12 (5.27)	3.91 (4.03)	0.098	0.140
BPI (mean ± SD) [a]	8.95 (12.46)	5.22 (8.07)	0.031	0.037
RBD-HK (mean ± SD) [a]	12.58 (17.09)	9.29 (13.53)	0.166	0.158
PDSS (mean ± SD) [a]	118.66 (22.52)	118.88 (24.8)	0.951	0.669
PDQ39 (mean ± SD) [a]	15.28 (17.06)	11.77 (14.55)	0.142	0.117
FSS (mean ± SD) [a]	23.91 (19.64)	19.68 (18.73)	0.128	0.169
ESS (mean ± SD) [a]	5.43 (6.01)	4.59 (5.45)	0.317	0.409
MDS-UPDRS (mean ± SD) [a]	43.69 (26.06)	44.73 (24.4)	0.801	0.802
PART I	8.15 (6.03)	7.16 (4.91)	0.296	0.291
PART II	10.2 (7.14)	10.43 (6.66)	0.837	0.992
PART III	25.62 (16.85)	26.43 (18.00)	0.763	0.733
NMSS (mean ± SD) [a]	28.17 (32.82)	21.09 (22.44)	0.118	0.131
cardiovascular	0.60 (2.00)	0.27 (0.77)	0.224	0.254
sleep	6.02 (7.47)	4.66 (6.03)	0.192	0.228
mood disorder	5.59 (10.47)	4.71 (9.03)	0.548	0.657
delusion	0.42 (2.26)	0.46 (2.21)	0.884	0.847
attention	2.29 (3.43)	1.34 (2.26)	0.044	0.046
gastrointestinal	2.89 (5.03)	2.25 (3.83)	0.362	0.388
urinary	4.44 (8.15)	3.11 (4.84)	0.233	0.230
sexual dysfunction	1.22 (4.41)	0.61 (2.58)	0.309	0.326
others	4.51 (6.15)	3.61 (6.00)	0.299	0.372
SCOPA-AUT (mean ± SD) [a]	8.52 (8.89)	6.13 (7.06)	0.053	0.064
gastrointestinal	2.83 (3.53)	1.98 (2.77)	0.084	0.088
urinary	3.14 (4.30)	2.39 (3.48)	0.210	0.289
cardiovascular	0.37 (1.00)	0.09 (0.35)	0.037	0.040
skin	1.56 (2.47)	1.29 (2.45)	0.427	0.505
sexual dysfunction	0.33 (1.23)	0.09 (0.55)	0.149	0.158
MMSE (mean ± SD) [b]	26.92 (3.12)	27.81 (3.06)	0.077	0.042
MoCA (mean ± SD) [b]	22.84 (4.64)	24.05 (5.09)	0.123	0.053

BPI, brief pain inventory; ESS, Epworth Sleepiness Scale; FSS, Fatigue severity scale; HAMA, Hamilton anxiety rating scale; HAMD, Hamilton depression rating scale; MDS, movement disorders society; MMSE, Mini-Mental State Examination; MoCA, Montreal Cognitive Assessment; NMSS, Non-Motor Symptoms Scale; PDQ-39, 39-item Parkinson's Disease Questionnaire; PDSS, Parkinson's disease sleep scale; RBD-HK, rapid eye movement sleep behavior disorder questionnaire - Hong Kong version; SCOPA-AUT, Scales for Outcomes in Parkinson's Disease-Autonomic questionnaire; SD, standard deviation; SS-16, Sniffin' Sticks 16; UPDRS, Unified Parkinson's Disease Rating Scale
a Hoehn Yahr Staging and gender were taken as adjustment
b Hoehn Yahr Staging, gender and education level were taken as adjustment

Table 3 the association between symptoms and genetic models of rs34778348

	Additive model	Dominant model			Dominant model (adjusted) [a]		
	p value	*p* value	OR	95% CI	*p* value	OR	95% CI
dysphagia [a]	0.455	0.502	0.74	(0.27, 1.67)	0.506	0.71	(0.24, 1.77)
sialorrhea [a]	0.049	0.066	0.55	(0.28, 1.02)	0.070	0.54	(0.26, 1.03)
symptom “full very quickly” [a]	0.298	0.237	1.63	(0.68, 3.53)	0.164	1.81	(0.74, 4.03)
constipation [a]	0.272	0.346	0.75	(0.40, 1.35)	0.539	0.82	(0.43, 1.52)
nocturia [a]	0.878	0.667	1.13	(0.65, 1.98)	0.435	1.27	(0.70, 2.33)
postural hypotension [a]	0.030	0.039	0.33	(0.10, 0.84)	0.050	0.35	(0.10, 0.89)
daytime sweatiness [a]	0.586	0.464	0.78	(0.38, 1.48)	0.639	0.85	(0.41, 1.64)
nocturnal sweatiness [a]	0.350	0.228	0.60	(0.24, 1.29)	0.276	0.63	(0.25, 1.37)
light sensitivity [a]	0.954	0.899	1.08	(0.25, 3.26)	0.880	1.10	(0.25, 3.35)
susceptible to cold [a]	0.358	0.401	0.70	(0.28, 1.51)	0.257	0.60	(0.22, 1.36)
susceptible to heat [a]	0.564	0.474	1.32	(0.58, 2.73)	0.558	1.28	(0.53, 2.76)
sexual dysfunction [a]	0.238	0.257	0.43	(0.07, 1.47)	0.323	0.48	(0.08, 1.66)
hallucination [a]	0.737	0.779	0.84	(0.20, 2.47)	0.577	0.65	(0.10, 2.37)
apathy [a]	0.484	0.339	0.68	(0.29, 1.42)	0.267	0.62	(0.25, 1.35)
pain [a]	0.458	0.405	0.78	(0.43, 1.38)	0.356	0.75	(0.40, 1.37)
urination disorders (not nocturia) [a]	0.551	0.673	0.88	(0.48, 1.57)	0.766	0.91	(0.47, 1.69)
fatigue [a]	0.682	0.670	0.89	(0.51, 1.55)	0.707	0.89	(0.49, 1.62)
freezing of gait [a]	0.590	0.484	1.28	(0.62, 2.46)	0.402	1.38	(0.62, 2.85)
tremor [a]	0.555	0.759	0.92	(0.53, 1.61)	0.430	0.79	(0.43, 1.44)
hyposmia [a]	0.550	0.644	1.17	(0.62, 2.33)	0.688	1.15	(0.60, 2.32)
probable RBD [a]	0.455	0.502	0.74	(0.27, 1.67)	0.506	0.71	(0.24, 1.77)
	Overdominant model				Overdominant model (adjusted) [a]		
	p value	OR	95% CI		*p* value	OR	95% CI
dysphagia [a]	0.569	1.30	(0.57, 3.49)		0.554	1.35	(0.54, 4.11)
sialorrhea [a]	0.097	1.72	(0.93, 3.35)		0.098	1.77	(0.92, 3.61)
full very quickly [a]	0.196	0.58	(0.27, 1.41)		0.139	0.53	(0.24, 1.31)
constipation [a]	0.458	1.26	(0.70, 2.36)		0.671	1.15	(0.61,

						2.21)
nocturia [a]	0.478	0.81	(0.46, 1.44)	0.288	0.72	(0.39, 1.32)
postural hypotension [a]	0.048	2.87	(1.13, 9.68)	0.060	2.75	(1.07, 9.35)
daytime sweatiness [a]	0.366	1.38	(0.71, 2.89)	0.520	1.26	(0.64, 2.68)
nocturnal sweatiness [a]	0.148	1.92	(0.85, 5.13)	0.180	1.84	(0.81, 4.95)
light sensitivity [a]	0.846	0.88	(0.29, 3.83)	0.823	0.87	(0.29, 3.77)
susceptible to cold [a]	0.468	1.36	(0.63, 3.40)	0.307	1.59	(0.70, 4.30)
susceptible to heat [a]	0.402	0.72	(0.35, 1.64)	0.490	0.75	(0.35, 1.81)
sexual dysfunction [a]	0.282	2.22	(0.65, 13.91)	0.355	2.00	(0.57, 12.67)
hallucination [a]	0.832	1.14	(0.39, 4.89)	0.601	1.49	(0.41, 9.66)
apathy [a]	0.233	1.66	(0.77, 4.12)	0.173	1.86	(0.82, 5.00)
pain [a]	0.368	1.31	(0.73, 2.40)	0.321	1.37	(0.74, 2.59)
urination disorders (not nocturia) [a]	0.827	1.07	(0.59, 1.98)	0.914	1.04	(0.55, 1.99)
fatigue [a]	0.668	1.13	(0.64, 2.00)	0.707	1.12	(0.61, 2.05)
freezing of gait [a]	0.398	0.74	(0.38, 1.54)	0.349	0.70	(0.34, 1.55)
tremor [a]	0.992	1.00	(0.56, 1.76)	0.690	1.13	(0.61, 2.08)
hyposmia [a]	0.764	0.90	(0.45, 1.70)	0.844	0.93	(0.46, 1.80)
probable RBD [a]	0.465	1.28	(0.68, 2.55)	0.395	1.35	(0.69, 2.80)

CI: confidence interval; OR: odds ratio; RBD: Rapid eye movement sleep behavior disorder
a Hoehn – Yahr staging and gender were taken as adjustment

Table 4 the association between symptoms and haplotypes *LRRK2*

	rs11564148 A - rs34778348 G (n = 281)					
	p value	OR	95% CI	*p* value [a]	OR [a]	95% CI [a]
dysphagia [a]	0.442	1.24	(0.72, 2.14)	0.370	1.30	(0.73, 2.34)
sialorrhea [a]	0.738	0.94	(0.64, 1.38)	0.989	1.00	(0.67, 1.51)
full very quickly [a]	0.815	1.08	(0.57, 2.07)	0.562	1.22	(0.63, 2.37)
constipation [a]	0.022	0.64	(0.43, 0.94)	0.062	0.67	(0.45, 1.02)
nocturia [a]	0.087	0.72	(0.50, 1.05)	0.149	0.74	(0.50, 1.11)
postural hypotension [a]	0.610	0.88	(0.55, 1.42)	0.997	1.00	(0.61, 1.63)
daytime sweatiness [a]	0.711	0.92	(0.6,0 1.41)	0.960	0.99	(0.64, 1.54)
nocturnal sweatiness [a]	0.785	0.94	(0.58, 1.50)	0.958	0.99	(0.61, 1.60)
light sensitivity [a]	0.264	1.66	(0.70, 4.24)	0.194	1.81	(0.75, 4.63)
susceptible to cold [a]	0.319	0.78	(0.47, 1.28)	0.487	0.84	(0.5,0 1.38)
susceptible to heat [a]	0.245	0.72	(0.40, 1.26)	0.303	0.74	(0.41, 1.31)
sexual dysfunction [a]	0.952	0.98	(0.49, 1.97)	0.835	0.93	(0.45, 1.90)
hallucination [a]	0.834	0.92	(0.43, 2.00)	0.816	1.10	(0.48, 2.54)
apathy [a]	0.448	0.83	(0.52, 1.33)	0.668	0.90	(0.56, 1.46)
pain [a]	0.634	1.10	(0.75, 1.60)	0.300	1.23	(0.83, 1.84)
urination disorders (not nocturia) [a]	0.373	0.84	(0.57, 1.24)	0.685	0.92	(0.61, 1.39)
fatigue [a]	0.587	0.90	(0.62, 1.31)	0.888	1.03	(0.69, 1.53)
freezing of gait [a]	0.709	0.91	(0.56, 1.49)	0.786	1.08	(0.64, 1.82)
tremor [a]	0.533	1.13	(0.77, 1.64)	0.476	1.16	(0.77, 1.73)
hyposmia [a]	0.909	0.98	(0.64, 1.50)	0.855	0.96	(0.62, 1.49)
probable RBD [a]	0.334	0.82	(0.54, 1.23)	0.653	0.91	(0.59, 1.39)
	rs11564148 A - rs34778348 A (n = 54)					
	p value	OR	95% CI	*p* value [a]	OR [a]	95% CI [a]
dysphagia [a]	0.755	0.86	(0.31, 2.08)	0.755	0.85	(0.27, 2.23)
sialorrhea [a]	0.088	0.56	(0.28, 1.07)	0.116	0.57	(0.27, 1.13)
full very quickly [a]	0.204	1.78	(0.70, 4.19)	0.124	2.04	(0.78, 4.94)
constipation [a]	0.151	0.63	(0.33,	0.322	0.71	(0.36,

			1.17)			1.38)
nocturia [a]	0.929	1.03	(0.56, 1.87)	0.563	1.21	(0.64, 2.33)
postural hypotension [a]	0.021	0.24	(0.06, 0.69)	0.035	0.27	(0.06, 0.79)
daytime sweatiness [a]	0.497	0.78	(0.37, 1.55)	0.715	0.87	(0.41, 1.77)
nocturnal sweatiness [a]	0.256	0.61	(0.24, 1.36)	0.314	0.64	(0.25, 1.45)
light sensitivity [a]	0.555	1.51	(0.32, 5.42)	0.479	1.64	(0.35, 5.98)
susceptible to cold [a]	0.319	0.64	(0.25, 1.45)	0.258	0.59	(0.21, 1.39)
susceptible to heat [a]	0.698	1.17	(0.50, 2.56)	0.723	1.17	(0.47, 2.67)
sexual dysfunction [a]	0.287	0.44	(0.07, 1.61)	0.338	0.48	(0.07, 1.78)
hallucination [a]	0.785	0.84	(0.19, 2.68)	0.697	0.73	(0.11, 2.99)
apathy [a]	0.302	0.65	(0.27, 1.41)	0.304	0.63	(0.24, 1.44)
pain [a]	0.654	0.87	(0.46, 1.60)	0.751	0.90	(0.46, 1.72)
urination disorders (not nocturia) [a]	0.608	0.85	(0.44, 1.58)	0.814	0.92	(0.47, 1.78)
fatigue [a]	0.410	0.78	(0.42, 1.41)	0.533	0.82	(0.43, 1.55)
freezing of gait [a]	0.514	1.27	(0.59, 2.59)	0.325	1.50	(0.65, 3.29)
tremor [a]	0.757	0.91	(0.50, 1.66)	0.471	0.79	(0.42, 1.50)
hyposmia [a]	0.585	1.22	(0.61, 2.58)	0.640	1.19	(0.59, 2.55)
probable RBD [a]	0.202	0.63	(0.30, 1.25)	0.217	0.63	(0.29, 1.28)

CI: confidence interval; OR: odds ratio; RBD: Rapid eye movement sleep behavior disorder
reference haplotype: rs11564148 T - rs34778348 G (n = 214)
a Hoehn – Yahr staging and gender were taken as adjustment

Supplementary Files

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- SupplementaryTable3.docx
- supplementarytable2.docx
- SupplementaryTable1.docx