Review Article

Negative association of ubiquitin carboxyl-terminal esterase L1 (UCHL1) gene S18Y variant and risk of Parkinson’s disease: an updated meta-analysis

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Abstract: Ubiquitin carboxyl-terminal esterase L1 gene (UCHL1) has been considered inversely associated with Parkinson’s disease (PD). Fortunately, the S18Y variant of this gene has been demonstrated to have protective effects on PD among diverse populations, worldwide. The protective effects, however, have not been consistently replicated. To address this question, a meta-analysis was performed. A total of 20 studies, including 9,308 patients and 10,796 controls, were included in this meta-analysis. In a dominant genetic model, statistically significant inverse association of S18Y with PD, among the pooled group and Caucasians, was found (OR = 0.90, 95% CI = 0.83-0.97, \( P = 0.005 \); OR = 0.91, 95% CI = 0.85-0.98, \( P = 0.03 \)). A statistically significant OR, among the pooled group, was revealed in allele-based contract (OR = 0.94, 95% CI = 0.89-0.98, \( P = 0.007 \)). No obvious evidence for association was found, with a recessive model, in each group (non-Asians, Asians, and total). Y18 allele carrier rate was 17.58% in non-Asian subjects and 50.21% in Asian subjects (\( P < 0.00001 \)). The S18Y variant in UCHL1 genes was significantly associated with decreased risk of PD. However, reduced sensitivity of S18Y to PD was found in stratified analysis, though a low frequency of S18Y variant was observed in PD for both non-Asian and Asian populations. Further studies with larger sample sizes, especially in Asian populations, are warranted to confirm association between common UCHL1 variant S18Y and PD protection.

Keywords: Parkinson’s disease, Ser18Thr, UCHL1

Introduction

Parkinson’s disease (PD; MIM 168600) is the second most common neurodegenerative disease, having complex etiology. Combined effects of environmental factors and genetic susceptibility risks contribute to the pathogenesis of PD, with at least 18 disease-causing gene loci and 13 genes for Parkinson’s having been implicated [1, 2]. Lewy bodies (LBs), where ubiquitin, ubiquitin carboxyl-terminal esterase L1 (UCHL1; MIM 191342), proteosome subunits, and α-synuclein reside, are present in dopaminergic neurons of substantia nigra in the brain and are pathologic hallmarks of PD [3, 4]. UCHL1 genes, 11.5 Kb with 9 exons on chromosome 4 (4p14), encode UCHL1 which hydrolyzes small C-terminal adducts of ubiquitin to generate monomeric ubiquitin [5]. UCHL1, comprised of 1%–2% of soluble brain proteins, is abundant in all neurons and cell cytoplasm of the diffuse neuroendocrine system, testes, and ovaries [6]. Fortunately, S18Y, a variant (rs5030732) of the UCHL1 gene, has been demonstrated to have protective effects on sporadic PD, worldwide [2, 4, 7-9, 10-14], with a greater effect on the early-onset form of the disease [15, 16]. The protective effects, however, have not been consistently replicated [2, 17-21]. To clarify the protective effects of S18Y variants on PD, this updated meta-analysis was conducted, including all eligible studies published before November 1, 2013.

Material and methods

Data search strategy

Medline and Web of Science databases were searched using the following keywords:
Ubiquitin carboxyl-terminal esterase L1 gene S18Y variant and Parkinson's disease

Figure 1. Flow chart of selection and the details for exclusion.

Parkinson's disease OR Parkinson disease OR PD; S18Y OR Ser18Thr OR rs5030732; UCHL1 OR UCH-L1 OR UCH-L1 OR ubiquitin carboxyl-terminal esterase L1. An upper date limit of November 2013, with no lower date limit, was applied. Search data were for human beings and in English. In addition, references of eligible studies, related reviews within 5 years, and genetic association studies found in the PD Gene Database [http://www.pdgene.org] were manually retrieved.

Inclusion and exclusion criteria

Included studies met the following criteria: 1) Exploration of association between the UCHL1 gene S18Y variant and PD; 2) Case-control study or nested-case control study; 3) Provided the number of cases and controls with different genotypes; and 4) Distribution of genotypes among controls in Hardy-Weinberg equilibrium (HWE). Major exclusion criteria were: 1) Review, editorial, or comment; 2) Duplicated studies; and 3) Functional studies.

Data extraction

Two authors (S. Gu and Y. Liu) extracted data from all eligible studies, independently, using a standard form. Disagreements were resolved by discussion with co-authors. Data extraction included the following: first author's name, year of publication, ethnicity, country of origin, number of cases and controls, genotypes, alleles, and HWE in the control group, gene polymorphisms, and others. Data from a certain category not reported in the primary study was designated “-.” Ethnicities were categorized as Asians and non-Asians.

Statistical analyses

Odds ratios (OR) with 95% CIs were used to determine strength of association between S18Y variant in UCHL1 genes and patients with PD risk. Chi-square goodness of fit was used to test deviation from HWE for control groups of each study. Samples with P values below 0.05 were considered out of HWE and were excluded from this meta-analysis. This study defined pooled OR, which are OR1, OR2, and OR3 for AA vs CC, CA vs CC, and AA vs CA, respectively. The biological justification for choice of the genetic model was determined by multiple pairwise comparison. Autosomal recessive (AA vs CA plus CC), dominant (AA plus CA vs CC), additive (AA plus CA vs CA plus CC), and complete over-dominant (AA plus CC vs CA) were examined. Heterogeneity between studies was assessed using Cochran's Q test and Higgins I². P values of less than 0.10 indicated heterogeneity to be statistically significant and I² values more than 50% indicated higher levels of heterogeneity. If heterogeneity existed among these studies, the random effects model (Der Simonian and Laird method) was used, otherwise, fixed-effects model (Mantel-Haenszel method) was applied. Subgroup analyses were performed by ethnicity.

One-way sensitivity analysis was performed to determine the stability of results and each individual study in the meta-analysis was omitted to reflect the influence of individual datasets on pooled OR [22]. Visual inspection of funnel plots and Peter's test statistics were used to assess presence of publication bias. P-values below 0.05 were considered significantly different. Cumulative meta-analysis and recursive
Table 1. Descriptive and clinical characteristics of 24 studies on \textit{UCHL1} S18Y variant and PD: 10,263 cases and 11,484 controls

<table>
<thead>
<tr>
<th>First author-year</th>
<th>Ethnicity</th>
<th>Country of Origin</th>
<th>Enroll-ment source</th>
<th>Type of PD</th>
<th>Case (n = 9382)</th>
<th>Controls (n = 10951)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Wang L-2011</td>
<td>Asian</td>
<td>China</td>
<td>Clinic-based</td>
<td>Sporadic</td>
<td>408 (56.9)</td>
<td>398 (54.5)</td>
</tr>
<tr>
<td>Wu YR-2010</td>
<td>Asian</td>
<td>Taiwan</td>
<td>Clinic-based</td>
<td>Sporadic</td>
<td>517 (56.3)</td>
<td>518 (52.1)</td>
</tr>
<tr>
<td>Zhang ZJ-2008</td>
<td>Asian</td>
<td>China</td>
<td>Clinic-based</td>
<td>Sporadic</td>
<td>600 (59.0)</td>
<td>334 (53.9)</td>
</tr>
<tr>
<td>Tan EK-2006</td>
<td>Asian</td>
<td>Singapore</td>
<td>Clinic-based</td>
<td>Sporadic</td>
<td>335 (56.4)</td>
<td>341 (53.4)</td>
</tr>
<tr>
<td>Wang J-2002</td>
<td>Asian</td>
<td>China</td>
<td>Clinic-based</td>
<td>Sporadic</td>
<td>160 (55.6)</td>
<td>160 (55.6)</td>
</tr>
<tr>
<td>Miyake Y-2012</td>
<td>Asian</td>
<td>Japan</td>
<td>Clinic-based</td>
<td>Sporadic</td>
<td>229 (38.4)</td>
<td>357 (38.9)</td>
</tr>
<tr>
<td>Snapinn KW-2011</td>
<td>Asian</td>
<td>Japan</td>
<td>Clinic-based</td>
<td>Sporadic</td>
<td>605 (42.6)</td>
<td>1620 (45.2)</td>
</tr>
<tr>
<td>Toda T-2003</td>
<td>Asian</td>
<td>Japan</td>
<td>Clinic-based</td>
<td>Sporadic</td>
<td>232 (49.1)</td>
<td>249 (51.0)</td>
</tr>
<tr>
<td>Satoh J-2001</td>
<td>Asian</td>
<td>Japan</td>
<td>Clinic-based</td>
<td>Sporadic</td>
<td>74 (37.8)</td>
<td>155 (51.0)</td>
</tr>
<tr>
<td>Zhang J-2000</td>
<td>Asian</td>
<td>Japan</td>
<td>Clinic-based</td>
<td>Sporadic</td>
<td>160 (55.4)</td>
<td>160 (58.1)</td>
</tr>
<tr>
<td>Xiromerisiou G-2011</td>
<td>Caucasian</td>
<td>Greece</td>
<td>-</td>
<td>Sporadic</td>
<td>399 (57.6)</td>
<td>420 (54.5)</td>
</tr>
<tr>
<td>Carmine Belin A-2007</td>
<td>Caucasian</td>
<td>Sweden</td>
<td>Clinic-based</td>
<td>Sporadic</td>
<td>296 (60.0)</td>
<td>235 (57.0)</td>
</tr>
<tr>
<td>Healy DG-2006</td>
<td>Caucasian</td>
<td>Europe</td>
<td>Clinic-based</td>
<td>Sporadic</td>
<td>1536 (58.3)</td>
<td>1487 (56.1)</td>
</tr>
<tr>
<td>Elbaz A-2003</td>
<td>Caucasian</td>
<td>France</td>
<td>Community-based</td>
<td>Sporadic</td>
<td>209 (56.9)</td>
<td>488 (59.2)</td>
</tr>
<tr>
<td>Leveque C-2001</td>
<td>Caucasian</td>
<td>France</td>
<td>-</td>
<td>Familial</td>
<td>114 (47.4)</td>
<td>93 (49.5)</td>
</tr>
<tr>
<td>Savettieri G-2001</td>
<td>Caucasian</td>
<td>Italy</td>
<td>-</td>
<td>Sporadic</td>
<td>169 (58.6)</td>
<td>165 (45.5)</td>
</tr>
</tbody>
</table>

**Enrollment source and Genotype/allele**

Case and controls with Clinic-based enrollment source are based on the UKP-DBB and Calne criteria. HWE (P) values are given for controls only.
<table>
<thead>
<tr>
<th>Study Source</th>
<th>Ethnicity</th>
<th>Country</th>
<th>Study Type</th>
<th>Sample Size</th>
<th>Age Median (Years)</th>
<th>Duration of Disease (Years)</th>
<th>Cases</th>
<th>Controls</th>
<th>Odds Ratio</th>
<th>95% CI</th>
<th>P-Value</th>
</tr>
</thead>
<tbody>
<tr>
<td>Mellick et al.</td>
<td>Caucasian</td>
<td>Australia</td>
<td>Clinic-based Sporadic</td>
<td>142</td>
<td>48.6</td>
<td>66.0</td>
<td>1992</td>
<td>100</td>
<td>33</td>
<td>9</td>
<td>233</td>
</tr>
<tr>
<td>Facheris et al.</td>
<td>Caucasian</td>
<td>USA</td>
<td>Clinic-based Sporadic</td>
<td>-</td>
<td>406</td>
<td>43.3</td>
<td>-</td>
<td>-</td>
<td>1992</td>
<td>272</td>
<td>125</td>
</tr>
<tr>
<td>Zhang et al.</td>
<td>Caucasian</td>
<td>Australia</td>
<td>Clinic-based Sporadic</td>
<td>153</td>
<td>-</td>
<td>60.6</td>
<td>-</td>
<td>1992</td>
<td>108</td>
<td>40</td>
<td>5</td>
</tr>
<tr>
<td>Chung et al.</td>
<td>Caucasian</td>
<td>USA</td>
<td>Clinic-based Sporadic</td>
<td>1103</td>
<td>64.1</td>
<td>62.2</td>
<td>68</td>
<td>-</td>
<td>749</td>
<td>323</td>
<td>29</td>
</tr>
<tr>
<td>Hutter et al.</td>
<td>Caucasian</td>
<td>USA</td>
<td>Clinic-based Sporadic</td>
<td>1757</td>
<td>67.7</td>
<td>58.7</td>
<td>68</td>
<td>-</td>
<td>1191</td>
<td>509</td>
<td>57</td>
</tr>
<tr>
<td>Maraganore et al.</td>
<td>Caucasian</td>
<td>USA</td>
<td>Clinic-based Sporadic</td>
<td>-</td>
<td>298</td>
<td>62.1</td>
<td>-</td>
<td>-</td>
<td>-</td>
<td>-</td>
<td>-</td>
</tr>
</tbody>
</table>

*NB: The table above represents a summary of studies investigating the association between the Ubiquitin carboxyl-terminal esterase L1 gene S18Y variant and Parkinson's disease.*
cumulative meta-analysis were used to estimate whether summary OR changed over time as more data accumulated.

All analyses were performed using Review Manager software (version 5.1.2, Cochrane Collaboration, Software Update, Oxford, United Kingdom).

Results

Literature collection

As shown in Figure 1, 89 articles on S18Y variant and PD risk were included in the initial search. A total of 22 articles were obtained after removal of duplicated publications [2-4, 7, 8, 11-27]. Principal characteristics of these studies are summarized in Table 1. Two studies [4, 21], significantly apart from HWE in controls, were excluded (P = 0.0009, P = 0.015; Table 1).

Finally, 9,308 patients and 10,796 unrelated controls, from 8 groups of Asians and 13 groups of non-Asians, were included in the current meta-analysis (Figure 2). Sample sizes ranged from 207 to 3,773. Controls were primarily healthy populations and matched patients in age and ethnicity (Figure 3). Y18 allele carrier rates were 17.58% in non-Asian control subjects and 50.21% in Asian control subjects (P < 0.00001).

Quantitative data synthesis

Estimated OR1, OR2, and OR3 were 0.88 (95% CI = 0.79-0.99), 0.91 (95% CI = 0.85-0.98), and 0.99 (95% CI = 0.91-1.08). Wald tests indicated that OR1 and OR2 were significant (P < 0.05) while OR3 was not significant (P = 0.82). The genetic model was mostly likely to be dominant (Table 2).

Meta-analysis results

Moderate degrees of inconsistencies were found in this present meta-analysis, but no evidence of statistical heterogeneity was observed (All 0% < I² < 50%). First, allele-based meta-analysis was performed. Overall OR, under a fixed effects model for allele-based contrast, was 0.94 (95% CI = 0.90-0.99) and statistically significant (P = 0.013). As suggested by the dominant model, strong association between the S18Y variant and combined PD was observed (ORs = 0.91, 95% CI = 0.85-0.97, P = 0.005). To determine overall gene effects, analysis of all genetic models (autosomal recessive, dominant, complete over-dominant, and addi-

Figure 2. Forest plot (fixed effects model) of PD risk associated with UCHL1 S18Y variant for allele-based contrast.
The study aimed to evaluate the association between the S18Y variant of the UCHL1 gene and Parkinson's disease (PD) using a meta-analysis of 20 studies. The meta-analysis included 10,796 patients and 9,308 controls. The results showed a significant decrease in the risk of PD associated with the S18Y variant under the dominant model (OR = 0.908, 95% CI 0.847-0.966, P = 0.003). The analysis also revealed ethnic differences, with a significant decrease in the risk of PD in non-Asians (OR = 0.903, 95% CI = 0.831-0.981, P = 0.015) but no significant association in Asians (OR = 1.00, 95% CI = 0.88-1.13, P = 0.43) by fixed effects meta-analysis (existed heterogeneity with I² = 1.2%, P = 0.43) (Table 3, Figures 2, 3).

**Sensitivity analyses**

Upon omission of each individual study, corresponding pooled ORs were not altered materially (data not shown).

**Publication bias**

Begg's funnel plot and Egger's test were performed to derive the publication bias of this meta-analysis. The funnel plot suggested no presence of publication bias. Moreover, the P-value of Begg's test was 0.31 and P-value of Egger's test was 0.32 for the dominant model, providing statistical evidence for funnel plot symmetry. No presence of publication bias was found in the recessive genetic model or allele-based contrast.

**Discussion**

To clarify roles of the S18Y variant of UCHL1 genes in PD, the most comprehensive meta-analysis of 20 studies was conducted, to date, including 10,796 patients and 9,308 unrelated controls. This present meta-analysis demonstrated that S18Y had significantly reverse correlation with PD, in overall comparisons, under the dominant inheritance model (OR = 0.908, 95% CI 0.847-0.966, P = 0.003). Considering that potential ethnic differences might be associated with distribution of genotypes, subgroup analysis by ethnicity of study population was also performed, obtaining similar results to that of overall studies in Caucasians (OR = 0.903, 95% CI = 0.831-0.981, P = 0.015). Although association in one previous meta-analysis was...
Ubiquitin carboxyl-terminal esterase L1 gene S18Y variant and Parkinson’s disease

Table 2. Data notation for dichotomous outcome

<table>
<thead>
<tr>
<th></th>
<th>OR$_1$</th>
<th>OR$_2$</th>
<th>OR$_3$</th>
</tr>
</thead>
<tbody>
<tr>
<td>Overall</td>
<td>0.883 (0.786-0.992)</td>
<td>0.912 (0.850-0.979)</td>
<td>0.990 (0.906-1.082)</td>
</tr>
<tr>
<td>Asian</td>
<td>0.89 (0.77-1.03)</td>
<td>0.93 (0.82-1.05)</td>
<td>0.96 (0.84-1.08)</td>
</tr>
<tr>
<td>Non-Asian</td>
<td>0.87 (0.72-1.06)</td>
<td>0.90 (0.83-0.99)</td>
<td>1.03 (0.91-1.17)</td>
</tr>
</tbody>
</table>

OR$_1$: AA vs CC; OR$_2$: CA vs CC; OR$_3$: AA vs CA.

Table 3. Meta-analysis of S18Y polymorphism and risk of PD in each subgroup

<table>
<thead>
<tr>
<th>Sample size (case/control)</th>
<th>Dominant OR (95% CI)</th>
<th>P</th>
<th>Recessive OR (95% CI)</th>
<th>P</th>
<th>Allele OR (95% CI)</th>
<th>P</th>
</tr>
</thead>
<tbody>
<tr>
<td>Overall</td>
<td>0.91 [0.85-0.97]</td>
<td>0.005</td>
<td>0.96 [0.88-1.05]</td>
<td>0.14</td>
<td>0.94 [0.90-0.99]</td>
<td>0.013</td>
</tr>
<tr>
<td>Asian</td>
<td>0.92 [0.81-1.03]</td>
<td>0.14</td>
<td>0.88 [0.72-1.08]</td>
<td>0.10</td>
<td>0.94 [0.88-1.01]</td>
<td>0.11</td>
</tr>
<tr>
<td>Non-Asian</td>
<td>0.90 [0.83-0.98]</td>
<td>0.015</td>
<td>1.00 [0.88-1.13]</td>
<td>0.43</td>
<td>0.94 [0.88-1.00]</td>
<td>0.059</td>
</tr>
</tbody>
</table>

Figure 4. Forest plot (fixed effects model) of PD risk associated with UCHL1 S18Y variant for recessive genetic model.

significant under a recessive model in Asian populations, this present study showed a null result under any genetic model [9].

UCHL1, alternatively designated protein gene product 9.5 (PGP9.5), is a deubiquitinating enzyme which hydrolyzes small adducts of ubiquitin and generates free monomeric ubiquitin form ubiquitin proteins [4]. It has been demonstrated that UCHL1 protein possesses two opposing enzymatic activities affecting alpha synuclein degradation [23]. UCHL1 hydrolyzes poly-ubiquitin chains, promoting the ubiquitination and proteasomal degradation of alpha-synuclein in its monomeric form [24]. However, UCHL1 also exhibits a second dimerization-dependent ubiquityl-ligase activity, as it ligates ubiquitin by means of a K63 linkage to alpha-synuclein, sparing it from proteasomal degradation [25]. The polymorphic S18Y variant of UCHL1 genes, associated with decreased PD risk, has reduced ligase activity but comparable hydrolase activity to the wild-type enzyme [12].
There are several reasons why UCHL1 genes may play a role in the etiology of PD. First, it is one of the most abundant proteins in the brain, representing up to 2% of total soluble brain protein. Additionally, its function of ligase activity and hydrolase activity of UCHL1 may play a role in proteasomal protein degradation in the brain, a critical process for neuronal health [5]. Second, it is found in Lewy bodies, one of the pathological hallmarks of PD [26]. Third, UCHL1 is intimately involved with the ubiquitin-dependent proteolytic system, a pathway strongly implicated in mechanisms of inclusion body formation and neurodegeneration [17]. Fourth, mice lacking UCHL1 genes develop neurodegenerative disorders [17]. Fifth, downregulation and extensive oxidative modification of UCHL1 have been observed in the brains of Alzheimer’s disease and PD patients [5]. Another possible mechanism may be that the presence of 18Tyr allele in UCHL1 genes simply represents a linkage disequilibrium with undefined genetic variations that play a major role in protecting against development of PD, presumably by upregulating production of neurotrophic factors or downregulating oxidative stress-inducing stimuli [4].

UCHL1 proteins have three catalytically active sites, composed of Cys90, His161, and Asp176, in addition to an oxyanion hole residue at Gln84 [27]. However, the Ser18Tyr substitution is unlikely to produce a discernible change in the secondary structure of UCHL1 proteins or modify its N-myristoylation sites, or a series of phosphorylation sites mediated by protein kinase C and casein kinase II [4].

There are some limitation to this meta-analysis. First, heterogeneity existed in the results of this meta-analysis. Although this likelihood was minimized by performing a careful search of published studies, using explicit criteria for study inclusion and performing strict data extraction and analysis, significant inter-study heterogeneity, nevertheless, existed in nearly every comparison. Second, sample sizes in most of the included studies was small, increasing the probability of false positives or false negatives. Third, different diagnostic criteria, different study designs, and control selections contributed to the possibility of false-positive results. Furthermore, negative small studies remained unpublished, contributing to publication bias [28]. In conclusion, the results of this present meta-analysis have provided evidence that S18Y variant is a protective factor in determining susceptibility to PD. The effects of this variant were not identified by subgroup analysis, stratified by ethnicity. Given that identicalness was found in each ethnicity, more studies including stratified analysis by ethnicity should be carried out to increase sensibility. Most importantly, further functional studies of the S18Y variant, in both cell and animal models, would be of great interest and benefit.

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Disclosure of conflict of interest

None.

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References

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