Published in final edited form as:
Parkinsonism Relat Disord. 2009 July ; 15(6): 466–467. doi:10.1016/j.parkreldis.2008.09.001.

Haplotype analysis of Lrrk2 R1441H carriers with parkinsonism

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Abstract

The Roc domain of the Lrrk2 protein harbors two pathogenic mutations which cause autosomal dominant parkinsonism (R1441C and R1441G). A third putatively pathogenic variant (R1441H) has been identified in four probands of diverse ethnicity with parkinsonism. Herein we show that the R1441H substitutions lie on different haplotypes within our patients, confirming this codon as a mutational hotspot. The absence of this variant in control subjects and the presence of two other pathogenic variants at this amino acid position collectively support the contention that R1441H is a pathogenic substitution.

Keywords

Parkinson's disease; Leucine-rich repeat kinase 2; R1441H

Introduction

Leucine-rich repeat kinase 2 (*LRRK2*) variants cause familial and sporadic parkinsonism. Pathogenic substitutions are distributed across different domains of the Lrrk2 protein; Ras of complex proteins (Roc; R1441G/C), C-terminal of Roc (COR; Y1699C) and mitogen-activated protein kinase kinase kinase (MAPKKK G2019S & I2020T), and recently, substitutions in the COR (R1628P) and WD40 domain (G2385R) have been associated with disease-risk in ethnic Chinese patients [1–3].

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The Roc domain contains two confirmed and one putative pathogenic variant at a single codon, arginine (R)1441. The original descriptions of Lrrk2 substitutions in parkinsonism described R1441C (*LRRK2* 4321C>T) and R1441G (*LRRK2* 4321C>G), and subsequent studies identified R1441H (*LRRK2* 4322G>A) [4,5]. Although, the R1441G substitution appears to be geographically restricted to Northern Spain with evidence of a common founder, R1441C appears to be worldwide and have occurred as at least three independent mutational events [6,7].

Four families with a history of parkinsonism have been identified to harbor R1441H [4,5,8, 9], and given its amino acid position and absence in over 3500 controls screened in studies to date, it is most likely the cause of their disease. However, the families are not large enough to demonstrate definitive co-segregation with disease. The four families are of diverse ethnicity (Portuguese, Greek, US and Taiwanese) and do not share a known ancestral lineage. This study set out to infer haplotypic structure for the chromosomal 12q12 region flanking *LRRK2* in R1441H carriers to establish if there is any evidence of a founder-effect.

Subject and Methods

Parkinsonism was diagnosed by movement disorder neurologists, according to published criteria and family members of the index case were examined if available [10]. The institutional review boards at each institution approved the study and each participant provided signed informed consent. Genomic DNA was extracted from peripheral blood using standard protocols. Direct sequencing of exon 31 was used to verify *LRRK2* 4322G>A (R1441H) mutation carrier status. For the Taiwanese and US Lrrk2 R1441H carriers all 51 exons of the *LRRK2* gene were sequenced with no other putative pathogenic variants observed. Adjacent genetic markers (14 single nucleotide polymorphisms (SNPs) and eight microsatellites) spanning ~6 Mb across the *LRRK2* locus were selected to infer haplotype structure in families harboring the Lrrk2 R1441H substitution as previously described [7]. All PCR primers and conditions are available on request.

Results and Discussion

The inferred haplotype data suggest that the R1441H substitution has arisen on multiple independent occasions (Table). The haplotypes of the R1441H carriers of European descent show diversity which may indicate a number of independent founders (Table). Even though it appears the R1441H carriers do not have a single common founder the clinical presentation of affected carriers appears to be similar to typical Parkinson's disease with an age at onset range of 32–64 years (median 54.5 years). All initially display levodopa responsive parkinsonism, however disease in one of the siblings from the Greek R1441H family appeared to transition into a progressive supranuclear palsy-like disorder [9]. These observations are reminiscent of one patient from a family with the Lrrk2 R1441C mutation (Family D), who displayed predominant tau rather than a-synuclein pathology, on post-mortem examination [7]. Neuropathologic studies in Lrrk2 R1441H carriers may provide further insight.

The occurrence of Lrrk2 R1441H in four kindreds, the absence in >3500 healthy controls and the pathogenicity of other variants occurring at this amino acid (R1441C and R1441G) collectively support the contention that R1441H is a pathogenic substitution. Longitudinal studies of these families may provide the necessary evidence for R1441H pathogenicity with disease co-segregation, and help elucidate the pathophysiology of Lrrk2 R1441H-associated parkinsonism.

Acknowledgments

We would like to thank all those who have contributed to our research. This work is supported by a Morris K. Udall Parkinson's Disease Research Center of Excellence (NINDS P50 #NS40256), the Department of Veterans Affairs (Merit Review Award), the Taiwan National Science Council (96-2628-B-002-103-MY2), by a grant from the "Internationaal Parkinson Fonds" (The Netherlands) to VB, and by a research grant from the "Parkinson Disease Foundation" to CS.

References

1. Di Fonzo A, Wu-Chou YH, Lu CS, van Doeselaar M, Simons EJ, Rohe CF, et al. A common missense variant in the LRRK2 gene, Gly2385Arg, associated with Parkinson's disease risk in Taiwan. Neurogenetics 2006;7(3):133–8. [PubMed: 16633828]
2. Mata IF, Wedemeyer WJ, Farrer MJ, Taylor JP, Gallo KA. LRRK2 in Parkinson's disease: protein domains and functional insights. Trends Neurosci 2006;29(5):286–93. [PubMed: 16616379]
3. Ross OA, Wu YR, Lee MC, Funayama M, Chen ML, Soto AI, et al. Analysis of Lrrk2 R1628P as a risk factor for Parkinson's disease. Ann Neurol 2008;64:88–92. [PubMed: 18412265]
4. Mata IF, Kachergus JM, Taylor JP, Lincoln S, Aasly J, Lynch T, et al. Lrrk2 pathogenic substitutions in Parkinson's disease. Neurogenetics 2005;6(4):171–7. [PubMed: 16172858]
5. Zabetian CP, Samii A, Mosley AD, Roberts JW, Leis BC, Yearout D, et al. A clinic-based study of the LRRK2 gene in Parkinson disease yields new mutations. Neurology 2005;65(5):741–4. [PubMed: 16157909]
6. Gonzalez-Fernandez MC, Lezcano E, Ross OA, Gomez-Esteban JC, Gomez-Busto F, Velasco F, et al. Lrrk2-associated parkinsonism is a major cause of disease in Northern Spain. Parkinsonism Relat Disord 2007;13(8):509–15. [PubMed: 17540608]
7. Haugarvoll K, Rademakers R, Kachergus JM, Nuytemans K, Ross OA, Gibson JM, et al. Lrrk2 R1441C parkinsonism is clinically similar to sporadic Parkinson disease. Neurology 2008;70(16 Pt 2):1456–60. [PubMed: 18337586]
8. Ferreira JJ, Guedes LC, Rosa MM, Coelho M, van Doeselaar M, Schweiger D, et al. High prevalence of LRRK2 mutations in familial and sporadic Parkinson's disease in Portugal. Mov Disord 2007;22 (8):1194–201. [PubMed: 17469194]
9. Spanaki C, Latsoudis H, Plaitakis A. LRRK2 mutations on Crete: R1441H associated with PD evolving to PSP. Neurology 2006;67(8):1518–9. [PubMed: 17060595]
10. Gelb DJ, Oliver E, Gilman S. Diagnostic criteria for Parkinson disease. Arch Neurol 1999;56:33–39. [PubMed: 9923759]

Table

Chromosome 12q12 haplotype analysis of Lrrk2 R1441H parkinsonism patients
LRRK2 4322 G>A (R1441H) mutation is highlighted in light grey.

LRRK2	NCBI 03/06	Genetic Marker*	Portuguese	Greek	US	Taiwanese
	33,305,718	**D12S2080**	184	184	192	192
	38,738,008	**D12S2194**	261	249	253	249
	38,873,924	**D12S2514**	291/294	291	294	294
ex5	38,918,058	**rs10878245**	T	C	T	T
int5	38,918,366	**rs10878246**	T	T/G	T	T
ex30	38,989,178	**rs7133914**	G	G	A	G
ex30	38,989,254	**rs11175964**	G	G	A	G
int30	38,989,339	**D12S2516**	254	252/254	254	252
int30	38,989,419	**rs11175966**	A	C	C	C
ex 31 R1441H	**38,990,504**	**rs34995376**	A	A	A	A
int33	39,000,026	**rs1896252**	T/C	T/C	C	T
ex34	39,000,101	**rs1427263**	C/A	C/A	A	C
ex34	39,000,140	**rs11176013**	A/G	A/G	G	A
ex34	39,000,168	**rs11564148**	T	T/A	T	T
int34	39,000,276	**rs11564205**	A	A	A	A
ex43	39,028,521	**rs10878405**	G	G/A	G	G
int43	39,028,630	**rs11176143**	G	A	G	G
int45	39,034,922	**D12S2518**	154	154/168	154	168
ex49	39,044,919	**rs3761863**	T/C	T/C	C	T/C
	39,116,885	**D12S2519**	140	132/140	140	138
	39,120,098	**D12S2520**	260	254/257	257	248
	39,128,754	**D12S2521**	319	363	315	327

* Microsatellite allele sizes were normalized using CEPH-control DNA (1331-01 and 1331-02) and approximate positions are determined from the NCBI March 2006 human genome assembly. Where phase is determined, only the genotype for the R1441H allele is shown. The United States (US) family report ancestry as from the United Kingdom.