Movement Disorders
CLINICAL PRACTICE

RESEARCH ARTICLE

Genetic Identification in Early Onset Parkinsonism among Norwegian Patients

Emil K. Gustavsson, MSc,[1,2,3,*] Joanne Trinh, BSc,[1] Marna McKenzie, BSc ,[1] Stephanie Bortnick, PhD,[1] Maria Skaalum Petersen, PhD,[4] Matthew J. Farrer, PhD,[1] Jan O. Aasly, MD[2,3]

Abstract: Background: An initial diagnosis of Parkinson's disease (PD) is challenging, especially in patients who have early onset and atypical disease. A genetic etiology for parkinsonism, when established, ends that diagnostic odyssey and may inform prognosis and therapy. The objective of this study was to elucidate the genetic etiology of parkinsonism in patients with early onset disease (age at onset <45 years).
Methods: Whole-exome sequencing, copy number variability, and short tandem repeat analyses were performed. The analyses were focused on genes previously implicated in parkinsonism and dystonia in patients with early onset parkinsonism. Genotype-phenotype correlations were assessed using regression models.
Results: The patient cohort was characterized by early onset, slowly progressive parkinsonism with a mean age at onset of 39.2 ± 5.0 years (n = 108). By 10 years of disease duration, the mean Hoehn & Yahr stage was 2.6 ± 0.8, the mean Unified Parkinson's Disease Rating Scale, part III (motor part) score was 24.9 ± 12.1 (n = 83), and 30 patients were cognitively impaired at the last examination (Montreal Cognitive Assessment score ≤ 26). Ten patients with typical early onset PD harbored homozygous or compound heterozygous mutations phosphatase and tensin homolog-induced putative kinase 1 (*PINK1*) (n = 4), parkin (*PRKN*) (n = 3), or the leucine-rich repeat kinase 2 (*LRRK2*) c.6055 G to A transition (n = 3). In addition, 5 patients with more atypical disease were compound heterozygotes for the glucocerebrosidase gene (*GBA*) (n = 3) 1 was heterozygous for solute carrier family 2, member 1 (*SLC2A1*) and another carried a novel ataxin 2 (*ATXN2*) exon 1 duplication. In most patients, the cumulative mutational burden did not appear to contribute to age at onset or progression
Conclusion: In this clinical series, 15 patients (14%) carried mutations that were linked to monogenic parkinsonism. *GBA* carriers were most likely to suffer an earlier cognitive demise. Nevertheless, the etiology for most patients with early onset PD remains to be determined.

Parkinsonism is characterized by resting tremor, rigidity, and bradykinesia,[1] and the most frequent form is Parkinson's disease (PD). This clinical syndrome is age-associated, neurodegenerative, and starts asymmetrically. Although these motor features respond well to dopamine-replacement therapy, several nonmotor symptoms, including autonomic dysfunction, hyposmia, pain, psychiatric and sleep disorders, cognitive dysfunction, and decline, remain problematic.[2–4] PD is generally late-onset (LOPD), affecting the elderly population (age at disease onset [AAO], >65 years), and only 4% of patients are diagnosed with

[1]Center for Applied Neurogenetics, Djavad Mowafaghian Center for Brain Health, Department of Medical Genetics, University of British Columbia, Vancouver, British Columbia, Canada; [2]Department of Neuroscience, Norwegian University of Science and Technology, Trondheim, Norway; [3]Department of Neurology, St. Olav's Hospital, Trondheim, Norway; [4]Department of Occupational Medicine and Public Health, The Faroese Hospital System, Torshavn, Faroe Islands

*Correspondence to: Dr. Emil K. Gustavsson, Djavad Mowafaghian Center for Brain Health, University of British Columbia, Vancouver, BC, Canada, V6T 2B5; E-mail:egustavsson@can.ubc.ca
Keywords: age at onset, early onset Parkinson's disease, mutation, parkinsonism.
Relevant disclosures and conflicts of interest are listed at the end of this article.
Supporting information may be found in the online version of this article.
The last two authors contributed equally to this work.
Received 9 February 2017; revised 26 March 2017; accepted 5 April 2017.
Published online 23 May 2017 in Wiley InterScience (www.interscience.wiley.com). DOI:10.1002/mdc3.12501

© 2017 International Parkinson and Movement Disorder Society

doi:10.1002/mdc3.12501

early onset PD (EOPD) (AAO, ≤45 years). A definitive diagnosis of PD is often challenging, because symptom onset is insidious; there is considerable clinical heterogeneity in presentation, progression, and response to therapy.[5] The accuracy of a clinical diagnosis of PD ranges between 76% and 92%, depending on the criteria used, stage of disease, and AAO.[6–11] Many patients with early onset forms of parkinsonism have dystonia, whereas many genetic forms of dystonias have parkinsonism.[12]

Although clinical evaluation and imaging help establish a diagnosis, genetic insights may be most insightful in defining clinical subtypes. Recessively inherited mutations in the phosphatase and tensin homolog-induced putative kinase 1 (*PINK1*) and parkin (*PRKN*) genes have been shown to cause a slowly progressive and mild early onset parkinsonism, with preservation of cognitive function and good response to levodopa (L-dopa) therapy.[13–15] In contrast, although recessively inherited mutations in adenosine 5′-triphosphatase 13A2 (*ATP13A2*); protein deglycase 1 DJ-1 (*DJ-1*); DnaJ (heat-shock protein 40) homolog, subfamily C, member 6 (*DNAJC6*); phospholipase A2 group VI (*PLA2G6*); F-box protein 7 (*FBXO7*); and synaptojanin 1 (*SYNJ1*) also result in juvenile/early onset parkinsonism, their presentation is more atypical, often including dystonia, spasticity, seizures, behavioral problems, and developmental delay, with little response to L-dopa.[16–19] Many other genes and genome-wide association loci and have been implicated in LOPD.[20]

Herein, we describe the genealogy and clinical characteristics of patients with EOPD originating in central Norway. Genome-wide genetic evaluation was performed, including whole-exome sequencing, copy number variation (CNV), and short tandem repeat analyses. Sequence and structural variability in genes implicated in early/juvenile onset parkinsonism, dystonia, and related neurological conditions are presented with their influence on AAO and clinical outcomes.

Materials and Methods

Patients

We studied 108 patients with early onset parkinsonism (AAO, 39.3 ± 5.0 years; range, 20–45 years; male:female (M:F) ratio, 1.6:1) originating from central Norway (patient characteristics are summarized in Table S1). All patients were examined and observed longitudinally by 1 movement disorder specialist (J.O.A.) at St. Olav's hospital in Trondheim, Norway, and were diagnosed according to UK Brain Bank criteria.[21] All participants provided informed consent before donating a blood sample for genetic analysis. Local ethics approval, which was independently reviewed by the University of British Columbia Research Ethics Board, was obtained.

Leucine-rich Repeat Kinase 2 p.G2019S and Short-tandem Nucleotide Repeat Screening

Leucine-rich repeat kinase 2 (*LRRK2*) c.6055 G to A (G > A) transition (p.G2019S) parkinsonism is a frequent cause of genetically determined PD in Central Norway[22]; therefore, all patients were screened for its presence using a TaqMan probe (Life Technologies, Carlsbad, CA).

Pathogenic short tandem nucleotide repeat expansions were examined in ataxin 2 (*ATXN2*); *ATXN3*; protein phosphatase 2, regulatory subunit B, β isoform (*PPP2R2B*); and TATA-box binding protein (*TBP*) using a fluorescent-labeled polymerase chain reaction primer with capillary electrophoresis on an ABI 3730xl Genome Analyzer (Thermo Fisher Scientific, Waltham, MA) and analyzed with Genemapper software (Life Technologies).

Whole-exome Sequencing and Variant Selection

Exonic regions were enriched using the Ion AmpliSeq exome kit (57.7 Mb) and sequenced on the Ion Proton (Life Technologies) with a minimum average coverage of 70 reads per base and an average read length of 150 bases. Reads were mapped to the National Center for Biotechnology Information Build 37.1 reference genome using the Ion Torrent Suite 5.0 (Thermo Fisher Scientific). Sequences with a mapping Phred quality score under 20, fewer than 10 reads, or over 95% strand bias were excluded from further analysis. Variants were subsequently annotated with ANNOVAR,[23] and allelic frequencies from the Exome Aggregation Consortium (ExAC) were included for the exclusion of frequently observed variants (minor allele frequency, >0.01). In addition, we employed Combined Annotation Dependent Depletion (CADD) analysis; CADD C-scores are an integrated metric of multiple annotations to rank functional, deleterious, and disease causal variants.[24] A C-score ≥ 20 indicates that the variant is predicted to be among the 1% most deleterious substitutions in the human genome and was employed as a cutoff to suggest those most likely to be causal of disease. All variants were validated by means of Sanger sequencing as previously described.[46] We analyzed variability in genes that were linked or associated with early onset parkinsonism, dystonia, or related neurologic conditions (Table 1).

Copy Number Analysis

Approximately 1.8 M single nucleotide polymorphisms were assessed genome-wide using an Illumina Multi-Ethnic Genome Array (Illumina, San Diego, CA). In addition to the patients, 165 neurologically healthy individuals from Scandinavia were included (Norway, n = 109; Faroe Islands, n = 55 l; age, 79.1 ± 9.0 years; age range, 66–100 years; M:F ratio, 0.7:1). The Illumina iScan Reader and GenomeStudio software were used to deduce genotype calls and CNVs from fluorescence intensity files (Illumina). Two algorithms were employed for the latter: CNVPartition version 3.2.0 (Illumina; using a default confidence threshold of 35, a minimum probe count of 3, and a minimal size window of 1 kb) and PennCNV.[25] Concordant findings were subsequently assessed. Copy number variants in *SNCA*, *PRKN*, and *PINK1* and in specific exons of *DJ-1*

doi:10.1002/mdc3.12501

TABLE 1 Major genes associated with early onset parkinsonism, dystonia, and related neurologic syndromes

Disease[a]	Inheritance	RefSeq gene	Position (hg19)	RefSeq
I. Early onset parkinsonism				
Juvenile-onset parkinsonism, Kufor-Rakeb syndrome; PARK9	AR	*ATP13A2*	chr1:17312453-17338423	NM_022089
Early onset parkinsonism; PARK7	AR	*DJ-1*	chr1:8021714-8045342	NM_007262
Juvenile-onset parkinsonism; PARK19	AR	*DNAJC6*	chr1:65775218-65881552	NM_001256864
Early onset parkinsonism; PARK15	AR	*FBXO7*	chr22:32870707-32894818	NM_012179
PD, Gaucher's disease	IC, Mu	*GBA*	chr1:155204239-155214653	NM_000157
Early onset parkinsonism; PARK6	AR	*PINK1*	chr1:20959948-20978004	NM_032409
Early onset parkinsonism, NBIA; PARK14	AR	*PLA2G6*	chr22:38507502-38577761	NM_003560
Early onset parkinsonism; PARK2	AR	*PRKN*	chr6:161768590-163148834	NM_004562
Early onset parkinsonism; PARK20	AR	*SYNJ1*	chr21:34001069-34100351	NM_003895
Early onset parkinsonism; PARK23	AR	*VPS13C*	chr15:62144590-62352664	NM_020821
II. Parkinson's disease				
PD; PARK22	AD	*CHCHD2*	chr7:56101569-56106576	NM_001320327
PD; PARK21	AD	*DNAJC13*	chr3:132136553-132257876	NM_015268
PD; PARK18	AD	*EIF4G1*	chr3:184032283-184053146	NM_182917
PD; PARK8	AD	*LRRK2*	chr12:40618813-40763086	NM_198578
PD; PARK1/4	AD	*SNCA*	chr4:90645250-90759447	NM_000345
PD; PARK17	AD	*VPS35*	chr16:46693589-46723144	NM_018206
III. Neurologic disease with parkinsonism				
Parkinsonism with spasticity	XLR	*ATP6AP2*	chrX:40440216-40465888	NM_005765
Wilson's disease	AR	*ATP7B*	chr13:52506806-52585630	NM_000053
Spinocerebellar ataxia 2; SCA2	AD	*ATXN2*	chr12:111890018-112037480	NM_002973
Spinocerebellar ataxia 3, Machado-Joseph disease; SCA3	AD	*ATXN3*	chr14:92524896-92572965	NM_004993
Perry syndrome/parkinsonism	AD	*DCTN1*	chr2:74588281-74607482	NM_004082
Progressive supranuclear palsy, FTD with parkinsonism	AR, AD	*MAPT*	chr17:43971748-44105699	NM_016835
Spinocerebellar ataxia 12; SCA12	AD	*PPP2R2B*	chr5:145969067-146258348	NM_181674
Intellectual disability with parkinsonism	XLR	*RAB39B*	chrX:155258241-155264589	NM_171998
Spinocerebellar ataxia 17; SCA17	AD	*TBP*	chr6:170863421-170881958	NM_003194
IV. Dystonia				
Rapid-onset dystonia-parkinsonism; DYT12	AD	*ATP1A3*	chr19:42470734-42498428	NM_152296
Dopa-responsive dystonia, Segawa syndrome; DYT5a	AR, AD	*GCH1*	chr14:55308724-55369542	NM_000161
Adult-onset cranial-cervical dystonia; DYT25	AD	*GNAL*	chr18:11689014-11885683	NM_182978
Paroxysmal nonkinesigenic dyskinesia; DYT8	AD	*PNKD*	chr2:219135115-219211516	NM_015488
Young-onset dystonia-parkinsonism; DYT16	AR	*PRKRA*	chr2:179296141-179315958	NM_003690
Paroxysmal kinesigenic dyskinesia; DYT10	AD	*PRRT2*	chr16:29823409-29827202	NM_145239
Myoclonic dystonia; DYT11	AD	*SGCE*	chr7:94214536-94285521	NM_001099401
GLUT1 deficiency syndrome; DYT18	AR, AD	*SLC2A1*	chr1:43391046-43424847	NM_006516
Dopa-responsive dystonia	AR	*SPR*	chr2:73114512-73119289	NM_003124
X-linked dystonia-parkinsonism; DYT3	XLR	*TAF1*	chrX:70586114-70685855	NM_004606
Dopa-responsive dystonia, Segawa syndrome; DYT5b	AR	*TH*	chr11:2185159-2193035	NM_199292
Adolescent-onset dystonia of mixed type; DYT6	AD	*THAP1*	chr8:42691817-42698474	NM_018105
Early onset generalized dystonia; DYT1	AD	*TOR1A*	chr9:132575221-132586441	NM_000113
"Non-DYT1" dystonia, whispering dysphonia; DYT4	AD	*TUBB4A*	chr19:6494330-6502330	NM_001289123

RefSeq, Reference Sequence database built by the National Center for Biotechnology Information; hg19, UC Santa Cruz Human Genome Browser-hg19 assembly; AR, autosomal recessive; NM, messenger RNA accession prefix; chr, chromosome; PD, Parkinson's disease; IC, isolated cases; Mu, multifactorial; NBIA, neurodegeneration with brain iron accumulation; AD, autosomal dominant; XLR, X-linked recessive; SCA, spinocerebellar ataxia/atrophy; FTD, frontotemporal dementia.
[a]Disease "numbers" indicate disease subtype according to the Online Mendelian Inheritance in Man database.

(exons 1, 3, 5, and 7), *ATP13A2* (exons 2 and 9); lipoprotein A (*LPA*) (exon 39), and tumor necrosis factor receptor superfamily member 9 (*TNFRSF9*) (exon 3) were confirmed using multiplex ligation-dependent probe amplification (MRC Holland, Amsterdam, the Netherlands; http://www.mlpa.com), and exonic deletions or multiplications were subsequently verified using quantitative polymerase chain reaction.

Statistical Analysis

Logistic regression models were used to assess mutation burden (e.g., the number of variants across candidate genes) on AAO, progression of motor symptoms after 10 years of disease duration, and cognitive decline at the last examination. Similar regression analyses were performed for specific genetic etiologies (e.g., *PRKN*, *PINK1*, glucocerebrosidase [*GBA*], and *LRRK2* carriers and EOPD).

Results

The cohort studied was characterized by early onset parkinsonism (mean AAO, 39.3 ± 5.0 years; range, 20–45 years; M:F ratio, 1.6:1; n = 108) with relatively slow motor symptom progression after 10 years of evaluation (mean Hoehn & Yahr [H&Y] stage, 2.6 ± 0.8; mean Unified Parkinson's Disease Rating Scale motor part [UPDRS III] score, 24.9 ± 12.1;

doi:10.1002/mdc3.12501

n = 83). No patients presented with the postural instability and gait difficulty (PIGD) phenotype at the time of study. In total, 83 patients (77%) developed dyskinesia after 6.2 ± 3.4 years (range, 0–14 years) of treatment, with an average L-dopa equivalent dosage (LEDD) of 688.4 ± 410.6 mg/day (range, 0–2200 mg/day). Forty-three patients (42%) underwent neurosurgical intervention (thalamotomy, globus pallidus internus, or subthalamic nucleus [STN] deep brain stimulation [DBS]) (Table S1). At the last examination (≥10 years after disease onset; n = 58), the majority of patients were cognitively spared; 26 patients had mild cognitive impairment (Montreal Cognitive Assessment [MoCA] scores, 18–26), three had moderate cognitive impairment (MoCA scores, 15–17), and 1 had dementia (MoCA score, 8). A family history of parkinsonism was reported in 50 patients (46.3%); 28 (25.9%) had an affected first-degree relative (20 with affected parents, and 11 with affected siblings), 20 (18.5%) had a second-degree relative, and 9 (8.3%) had a third-degree relative with parkinsonism (Fig. S2).

In total, 86 participants harbored 119 rare, nonsynonymous variants in 30 genes that have been implicated in parkinsonism and/or dystonia (Fig. 1, Table 1). However, there were no significant correlations ($P > 0.05$) with mutation burden and AAO ($r^2 < 0.0001$; n = 108), the severity of motor symptomatology (UPDRS score at 10 years of disease duration; $r = 0.0013$; n = 86), or cognitive impairment (MoCA score beyond 10 years of disease duration; $r^2 = 0.0648$; n = 58). There were 28 variants with CADD scores > 20, which were presumed to be most damaging to protein function (Table 2), and 14 structural variants were identified using multiethnic genome array and multiplex ligation-dependent probe amplification methods (Table 3).

We identified compound heterozygous or homozygous mutations in *PINK1* (n = 4) and *PRKN* (n = 3). Patient P-607 is a heterozygous carrier of a *PINK1* exon 4 and 5 deletion and a hemizygous *PINK1* c.1079 A>G (p.H360R) mutation in exon 5. Her mother (F35-1) is homozygous for the same *PINK1* exon 4 and 5 deletion. P-169 carries a heterozygous *PINK1* exon 4 and 5 deletion and a homozygous exon 1 c. 230T>C (p.L77P) mutation. P-1016 was heterozygous for a *PINK1* exon 4 duplication and was compound heterozygous for c.218C> T (p.S73L) and c.454C>T (p.R152W), with both parents asymptomatic at ages 50 and 57 years, respectively. P-029 carries a homozygous *PRKN* exon 4 deletion, with no known family history of disease. P-936 has a heterozygous *PRKN* exon 4 deletion and carries a heterozygous c.823C>T (p.R275W) mutation in exon 7. P-966 is a homozygous *PRKN* exon 3 and 4 deletion carrier. Sequence and structural variability in *PINK1* and *PRKN* is summarized in Tables 2 and 3. These patients collectively developed tremor and fluctuations early and had a typical, generally mild, L-dopa–responsive parkinsonism (mean H&Y stage, 2.9 ± 0.7; mean UPDRS III score, 27.5 ± 8.4; n = 5) without any cognitive impairment at their last evaluation.

In addition, we identified heterozygous mutations in *LRRK2* (n = 3) and *GBA* (n = 3) (Tables 2 and 4). *LRRK2* p.G2019S was identified in 3 patients (P-089, P-369, and P-824), and the former 2 have previously been reported.[22] They all developed tremor early and had a typical, generally mild, L-dopa–responsive parkinsonism (mean H&Y stage, 2.7 ± 0.6; mean UPDRS III score, 26.0 ± 15.:, n = 3). All three had STN DBS. Both P-369 and P-824 are cognitively spared, whereas P-089 now has moderate cognitive impairment (MoCA score, 15). We identified 3 compound heterozygote *GBA* carriers (P-221, P-261, and F78-1) who had the c.1226A>G (p.N370S) mutation, and none had any features of Gaucher's disease. In addition, P-221 is a carrier of a novel heterozygous c.1478G>A (p.G454D) mutation, whereas P-261 and his affected sister (F78-1) both carried a heterozygous c.1093G>A (p.E326K) variant. Both siblings also carry a pathogenic mutation for dystonia in torsin family 1 member A (*TOR1A*), namely, c.613T>A (p.F205I), albeit without this symptom. All 3 *GBA* carriers have STN DBS, which manages their parkinsonism (mean H&Y stage, 3.5 ± 0.7; mean UPDRS III score, 37.0 ± 8.5; n = 3) but have since developed cognitive impairment (MoCA score, 8–26) (Tables 3 and 4). In addition, 19 heterozygous carriers of *GBA* variants were identified, namely, c.1093G>A (p.E326K; n = 11), c.1223C>T (p.T369M; n = 6), c.1186T>A (p.W359R; n = 1), and c.595_597delinsC (p.L160fsTer1; n = 1) (Fig. 1).

One female patient (ES-117) is a heterozygous carrier of solute carrier family 2, member 1 (*SLC2A1*) c.805C>T (p.R269C). Her disease started with oculogyric crisis and early onset parkinsonism, which now more closely resembles an L-dopa–responsive dystonia (Tables 3 and 4). She has had symptoms for 19 years and is now severely affected, with an H&Y stage of 4 or 5. The mutation is de novo, because neither of her unaffected parents are carriers. One patient (P-459) was identified with a duplication of *ATXN2* exon 1, although his symptoms manifest as typical L-dopa–responsive PD. He has STN DBS and becomes akinetic-rigid every time his stimulator stops and the battery needs to be replaced. His son (DNA not available at this time) has the same disease presentation.

MoCA scores differed significantly between patients with and without pathogenic mutations (after adjusting for disease duration; $P = 0.010$), with the greatest difference observed between *GBA* and *PRKN* patients (−0.922 ± 0.443; $P = 0.037$). Otherwise, no significant differences were observed between AAO, H&Y stage, motor scores (UPDRS III), or known family history of parkinsonism ($P > 0.05$).

In addition to known disease-causing variability (Table 4), we identified an additional 13 "putatively pathogenic" sequence variants with minor allele frequencies < 0.01 and CADD scores > 20 (Table 2). We did not find any additional homozygous or compound heterozygous carriers of the genes implicated in recessively inherited disease or any repeat expansions in *ATXN2*, *ATXN3*, *PPP2R2B*, or *TBP*. Among the patients, and compared with control samples, there was no excess of CNVs (Table 3). Excluding the CNVs presented within *PINK1*, *PRKN*, and *ATXN2*, only 3 of the called CNVs were absent from controls, all of which were duplications (with 3 copy numbers). The carriers of CNVs in G protein subunit α L

doi:10.1002/mdc3.12501

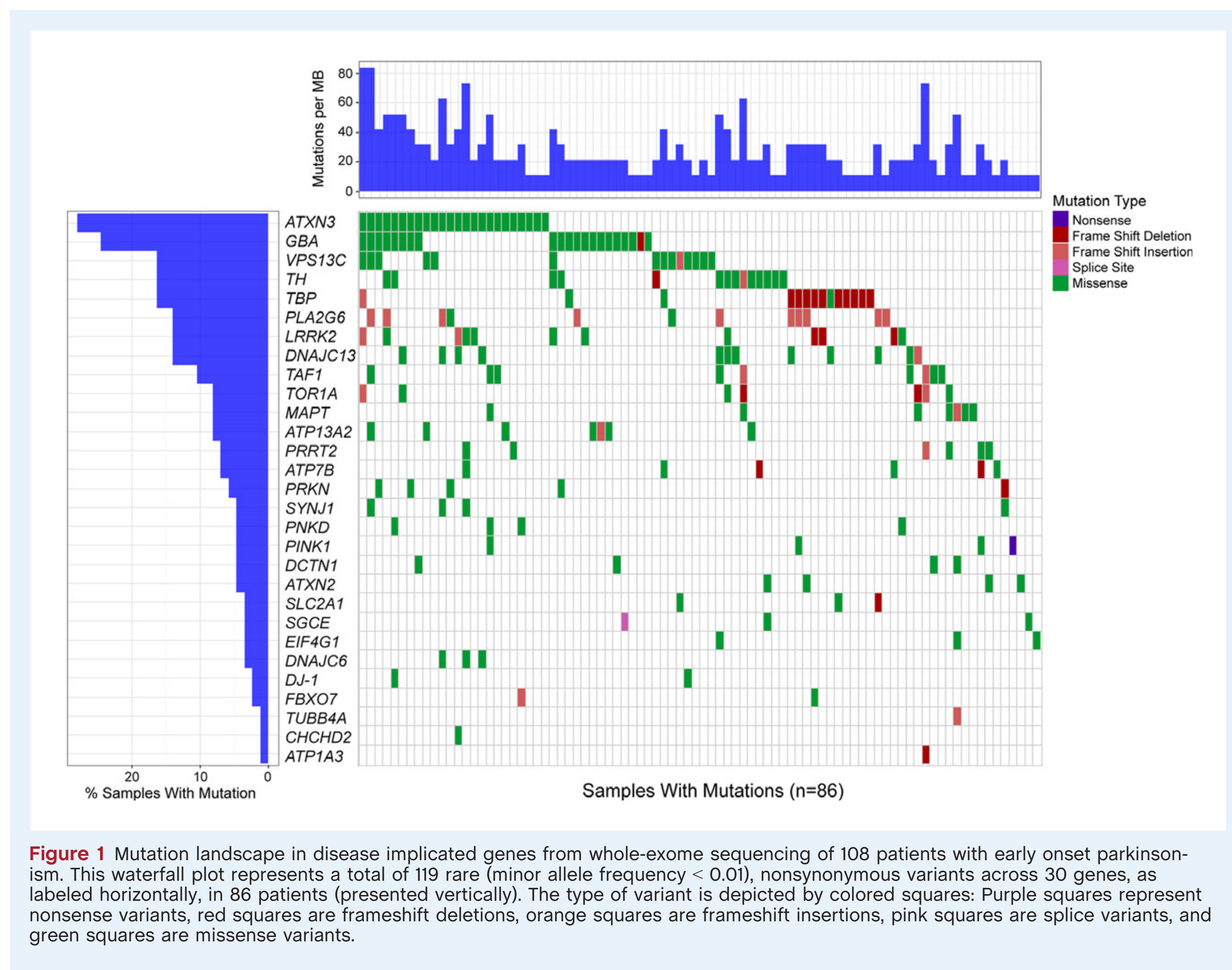

Figure 1 Mutation landscape in disease implicated genes from whole-exome sequencing of 108 patients with early onset parkinsonism. This waterfall plot represents a total of 119 rare (minor allele frequency < 0.01), nonsynonymous variants across 30 genes, as labeled horizontally, in 86 patients (presented vertically). The type of variant is depicted by colored squares: Purple squares represent nonsense variants, red squares are frameshift deletions, orange squares are frameshift insertions, pink squares are splice variants, and green squares are missense variants.

(*GNAL*) and paroxysmal nonkinesigenic dyskinesia (*PNKD*) do present with typical EOPD and no overlap with dystonia 8 (DYT8) (*PNKD*) or DYT25 (*GNAL*), suggesting the these CNVs are not the cause of their disease.

Discussion

In this study, we describe longitudinal clinical characteristics and comprehensive genetic analyses of 108 unrelated patients with EOPD (Table 1). The patients who were included are representative of those with early onset parkinsonism, although they were seen by a movement disorder specialist after referral by family physicians and general neurologists from the surrounding region. Although Central Norway maintains close-knit communities with relatively high kinship coefficients among seemingly unrelated individuals (data not shown), and the sample size is relatively small and subject to bias, the heritability of disease among affected individuals is considerable. Notably, 49 individuals (46.7%) reported a positive family history of parkinsonism versus 30.6% of patients with EOPD in Finland, and 10% to 14% of patients globally with LOPD.[26,27] Nevertheless, shared environmental factors may not be excluded.

We observed that 15 patients were carriers of previously described, disease-causing variability (Table 4), representing a diagnostic yield of 14%. Nevertheless, when susceptibility is attributed to a single, specific mutation(s), the outcomes in terms of AAO, motor and cognitive progression, and response to therapy appear to be complex. There is considerable heterogeneity in symptom onset and disease development among our participants. Importantly, 83 patients (86%) remain unexplained, for which, further investigation is warranted. Surprisingly, 86 of the 108 patients were carriers of 1 or more rare nonsynonymous variant(s) in a disease-causing gene. However, some variability may have been overlooked, because whole-exome sequencing does not fully cover all exonic or intronic regions. Nevertheless, accumulation of such variants does not appear to influence disease progression in terms of AAO, motor symptom severity, or cognitive function. This suggests that that disease outcomes in terms of AAO, progression, and cognitive impairment are more likely attributed to specific disease-causing mutations and/or environmental or stochastic factors.

doi:10.1002/mdc3.12501

TABLE 2 Rare, nonsynonymous, damaging variants (Combined Annotation Dependent Depletion score > 20) identified in genes associated with early onset parkinsonism, dystonia, and related syndromes

RefSeq gene	Position (hg19)	Nucleotide change	Amino acid change	rs ID	ExAC frequency	CADD score	No. of carriers
I. Early onset parkinsonism							
ATP13A2	Chr1:17319033	c.1793delinsGA	p.Q598RfsTer2	NA	NA	32	1 (Het)
FBXO7	Chr22:32894183	c.1235delinsTC	p.V412VfsTer2	NA	NA	23.6	1 (Het)
GBA	Chr1:155205634	c.1226A>G	p.N370S	rs76763715	2.20E-03	22.7	2 (Het)
	Chr1:155205013	c.1478G>A	p.G454D	NA	NA	24.8	1 (Het)
	Chr1:155206167	c.1093G>A	p.E326K	rs2230288	9.80E-03	20.33	12 (Het)
PINK1	Chr1:20960259	c.218C>T	p.S73L	rs202048763	2.82E-04	23.6	1 (Het)
	Chr1:20960271	c.230T>C	p.L77P	NA	NA	24.5	1 (Hom)
	Chr1:20964401	c.454C>T	p.R152W	rs45608139	2.47E-05	35	1 (Het)
	Chr1:20972172	c.1079A>G	p.H360R	NA	NA	25.3	1 (Het)
PLA2G6	Chr22:38531041	c.848A>G	p.D283G	NA	3.34E-05	28.5	1 (Het)
PRKN	Chr6:161771219	c.1310C>T	p.P437L	rs149953814	1.50E-03	25.5	1 (Het)
	Chr6:162206852	c.823C>T	p.R275W	rs34424986	2.10E-03	20.1	1 (Het)
	Chr6:162864410	c.101_103delinsG	p.Q34RfsTer5	NA	3.00E-04	23.6	1 (Het)
II. Parkinson's disease							
DNAJC13	Chr3:132247160	c.6509T>G	p.L2170W	rs140537885	2.30E-03	23	2 (Het)
	Chr3:132196983	c.2708G>A	p.R903K	rs141952333	2.00E-03	23.6	1 (Het)
LRRK2	Chr12:40734202	c.6055G>A	p.G2019S	rs34637584	3.87E-04	35	3 (Het)
	Chr12:40713856	c.4894G>C	p.E1632Q	rs200580973	NA	25.8	1 (Het)
III. Neurologic disease with parkinsonism							
ATXN2	Chr12:111893991	c.3586G>A	p.A1196T	rs369512638	3.30E-05	25.1	1 (Het)
	Chr12:111907995	c.3233C>T	p.A1078V	NA	4.12E-05	36	1 (Het)
MAPT	Chr17:44067341	c.1280C>T	p.S427F	rs143956882	1.50E-03	20.2	1 (Het)
TBP	Chr6:170878719	c.697G>A	p.A233T	NA	NA	26.8	1 (Het)
IV. Dystonia							
SGCE	Chr7:94259062	c.201T>A	p.F67L	NA	NA	26.7	1 (Het)
SLC2A1	Chr1:43395326	c.805C>T	p.R269C	rs200247956	2.85E-04	35	1 (Het)
TH	Chr11:2192979	c.38G>C	p.G13A	NA	NA	26.9	1 (Het)
	Chr11:2190989	c.296delinsAAAAAT	p.L99QfsTer3	NA	NA	24.5	1 (Het)
TOR1A	Chr9:132585071	c.229_233delinsT	p.I78KfsTer1	NA	NA	25.8	2 (Het)
	Chr9:132581031	c.613T>A	p.F205I	rs267607134	2.47E-05	36	1 (Het)
TUBB4A	Chr19:6495588	c.1075delinsTG	p.G308Ter	NA	NA	34	1 (Het)

RefSeq, Reference Sequence database built by the National Center for Biotechnology Information; hg19, UC Santa Cruz Human Genome Browser-hg19 assembly; rs ID, reference single nucleotide polymorphism number; ExAC, Exome Aggregation Consortium; CADD, Combined Annotation Dependent Depletion; Chr, chromosome; delins, deletion or insertion; NA, not applicable; Het, heterozygous; Hom, homozygous.

TABLE 3 Copy number variability within genes associated with early onset parkinsonism, dystonia, and related syndromes

RefSeq gene	Chromosomal band	Exons	Copy no.	No./total no. (%)	
				Affected carriers	Control carriers
I. Early onset parkinsonism					
PINK1	1p36.12	Ex4	3	1/105 (1.0)	1/165 (0.0)
		Ex4-5	0–1	3/105 (2.9)	0/165 (0.0)
PRKN	6q26	Ex3-4	0	1/105 (1.0)	0/165 (0.0)
		Ex4	1	1/105 (1.0)	0/165 (0.0)
		Ex5	0	1/105 (1.0)	0/165 (0.0)
GBA	1q21.3-q22	Complete gene	3	6/105 (5.7)	4/165 (2.4)
III. Neurologic disease with parkinsonism					
ATXN2	12q24.12	Ex1	3	1/105 (1.0)	0/165 (0.0)
IV. Dystonia					
ATP1A3	19q13.2	Complete gene	3	6/105 (7.6)	4/165 (2.4)
		Complete gene	1	0/105 (0.0)	1/165 (0.6)
GCH1	14q22.2	Ex1	3–4	4/105 (3.8)	0/165 (0.0)
		Ex1	1	0/105 (0.0)	1/165 (0.6)
GNAL	18p11.21	Ex1	3	1/105 (1.0)	0/165 (0.0)
PNKD	2q35	Complete gene	3	3/105 (1.9)	0/165 (0.0)
PRRT2	16p11.2	Complete gene	3	10/105 (9.5)	7/165 (4.2)
		Ex1-2	3	11/105 (10.5)	1/165 (0.6)
TUBB4A	19p13.3	Complete gene	3–4	4/105 (3.8)	2/165 (1.2)

RefSeq, Reference Sequence database built by the National Center for Biotechnology Information; hg19, UC Santa Cruz Human Genome Browser-hg19 assembly.

The clinical phenotype of *LRRK2*, *PINK1*, *PRKN*, *GBA*/*TOR1A*, and *ATXN2* carriers is that of typical L-dopa–responsive parkinsonism, of which 10 of 12 patients had a disease duration > 5 years (Table 4). *LRRK2* carriers are generally indistinguishable from patients who have typical PD, with variable AAO (range, 28–82 years).[28,29] Although onset is generally

doi:10.1002/mdc3.12501

TABLE 4 Clinical data from patients with early onset parkinsonism who carried pathogenic variability

Patient	RefSeq gene	Copy no. variant	Sequence variant	Sex	Age, y	AAO, y	UPDRS III score after 10 y	H&Y score after 10 y	Dyskinesia: Time after onset, y	LEDD	Surgery	MoCA score (years after onset)	Known family history of disease
P-607	*PINK1*	Ex4-5del (het)	p.H360R (hem)	Woman	71	43	19	3	8	650	NS	28 (15)	Yes
F35-1	*PINK1*	Ex4-5del (hom)	—	Woman	95	46	NA	2	0	200	Thalamotomy	27 (30)	Yes
P-169	*PINK1*	Ex4-5del (het)	p.L77P (hom)	Woman	74	44	37	3	7	1100	GPi DBS	28 (15)	Yes
P-1016	*PINK1*	Ex4dup (het)	p.S73L (het); p.R152W (het)	Woman	29	29	17	1	0	0	NS	28 (2)	No
P-029	*PRKN*	Ex4del (hom)	—	Man	65	35	22	2.5	4	2200	GPi DBS	30 (30)	No
P-936	*PRKN*	Ex4del (het)	p.R275W (het)	Woman	45	42	7	1	0	550	NS	28 (2)	No
P-966	*PRKN*	Ex3-4del (hom)	—	Man	77	30	32	4	14	600	STN DBS	29 (15)	Yes
P-221	*GBA*	—	p.N370S (het); p.G454D (het)	Woman	65	40	31	3	5	200	STN DBS	26 (15)	Yes
P-261	*GBA*	—	p.E326K (het); p.N370S (het)	Man	57	41	43	4	7	800	STN DBS	8 (15)	Yes
	TOR1A	—	p.F205I (het)										
F78-1	*GBA*	—	p.E326K (het); p.N370S (het)	Woman	63	50	32	2.5	8	300	STN DBS	20 (14)	Yes
	TOR1A	—	p.F205I (het)										
P-089	*LRRK2*	—	p.G2019S (het)	Man	63	43	44	3	4	300	STN DBS	15 (15)	Yes
P-369	*LRRK2*	—	p.G2019S (het)	Woman	62	43	18	3	4	300	STN DBS	26 (15)	Yes
P-824	*LRRK2*	—	p.G2019S (het)	Woman	48	40	16	2	2	450	STN DBS	27 (5)	No
ES-117	*SLC2A1*	—	p.R269C (het)	Woman	45	26	NA	4	5	400	NS	NA	No
P-459	*ATXN2*	Ex1dup (het)[a]	—	Man	60[b]	37	19	3	6	600	STN DBS	14 (20)	Yes

RefSeq, Reference Sequence database built by the National Center for Biotechnology Information; AAO, age at disease onset; AAS, age at surgery; del, deletion; dup, duplication; Ex, exon; H&Y, Hoehn & Yahr; LEDD, levodopa equivalent daily dose; het, heterozygous; hem, hemizygous; MoCA, Montreal Cognitive Assessment; NS, not surgery; hom, homozygous; GPi, globus pallidus internus (unilateral); DBS, deep brain stimulation; STN, subthalamic nucleus (bilateral).

[a]This copy number variation covers the previously reported pathogenic repeat expansion.

[b]The age at death is indicated.

doi:10.1002/mdc3.12501

after age 50 years, these patients all have early disease onset. It was demonstrated previously that the *LRRK2* p.G2019S variant in Norway stems from a common ancestor, which could explain the high frequency of carriers with an earlier disease phenotype.[30] Carriers of *LRRK2*, *PINK1*, *PRKN*, *GBA/TOR1A*, and *ATXN2* mutations benefit well from L-dopa and DBS with regard to their movement disorder and are suitable candidates for such therapeutic intervention.[31,32] However, cognitive performance in *GBA* carriers defined the most readily apparent clinical subtype, for which *GBA/TOR1A* carriers had the most severe and rapid decline, similar to previous reports.[33] Although the *GBA* p.E326K polymorphism does not cause disease, it has been suggested that it predicts a more rapid progression of cognitive impairment, as observed in these patients.[33] Nevertheless, no Gaucher's disease symptoms were manifest, although they were often noted in past reports.[34,35] We did not observe a significant difference in the frequency of disease-causing variability among patients with or without a known family history of parkinsonism (10 of 50 and 5 of 58 patients, respectively). However, we do not want to rule out this possibility only because it is difficult to examine relatives. In such instances, this information relies on the recollection of the patient and might not always be known, especially for a recessive disorder that generally does not manifest in every generation. Quantitative meta-analyses of larger EOPD cohorts are warranted to better define how different etiologies contribute to progression and treatment.

The *SLC2A1* p.R269C carrier in our study initially presented with parkinsonism, which subsequently progressed to an L-dopa–responsive, dystonic phenotype with early oculogyric crisis. The mutation appears to be de novo, which is a frequent occurrence for pathogenic mutations in *SLC2A1*.[36–38] Nevertheless, in infants, such mutations are more typical of glucose transporter type 1 deficiency syndrome (GLUT1-DS), which is associated with seizures (epilepsy). Only 10% have a nonepileptic form that includes a broad phenotypic spectrum of paroxysmal manifestations, intermittent ataxia, choreoathetosis, dystonia, parkinsonism, and alternating hemiplegia[39,40] Predicting the disease outcome of *SLC2A1* carriers is challenging; however, early implementation of a ketogenic diet may lessen the severity of multiple symptoms, including movement disorders.[41–44]

The patient who had an exon 1 duplication of *ATXN2* developed symptoms of parkinsonism at age 37 years. He has spinocerebellar ataxia type 2 (SCA2), which looks like L-dopa–responsive PD with a moderate cognitive impairment, and he had STN DBS many years ago. He still benefits from the surgery and deteriorates every time his battery needs to be replaced. *ATXN2* contains a polyglutamine tract located in exon 1, which is covered by the CNV duplication, in which long repeat expansions cause SCA2 and susceptibility to PD. However, to fully establish the pathogenicity of this CNV, further genotyping is warranted. In addition, whether this CNV and the damaging repeat expansions cause the same functional effect remains to be investigated.

Additional rare, putatively damaging variability was observed, although it not sufficient to cause recessively inherited, early onset parkinsonism. Both *DNAJC13* c.6509T>G (p.L2170W) and c.2708G>A (p.R903K) have been described as risk factors for parkinsonism in Norwegians.[44,45] *LRRK2* c.4894G>C (p.E1632Q) has not been described previously in any patients with parkinsonism, nor is it present in the ExAC database (version 0.3.1); however, 1 individual with chronic obstructive pulmonary disease has been described.[47] Microtubule-associated protein τ (*MAPT*) c.1280C>T (p.S427F), although rare, has been observed in 123 non-Finish Europeans from the ExAC database. Even considering incomplete penetrance, this is almost 4-fold higher than the expected number of individuals with early onset parkinsonism within that population. ε-Sarcoglycan (*SGCE*) c.201T>A (p.F67L) was identified in 1 patient who had parkinsonism starting at age 31 years and developed dyskinesia 1 year later. *SGCE* generally results in myoclonus dystonia with onset in childhood or early adolescence, without parkinsonism, suggesting that such variability might be a contributing factor to dyskinesia.[48] Inferences about genome-wide CNVs were not made beyond known risk loci. However, not including CNVs within *PINK1*, *PRKN*, and *ATXN2*, no other CNVs were found that would explain disease.

Genetic analysis of patients who initially present with early onset parkinsonism may assist in a differential diagnosis and in predicting disease prognosis and response to treatment. Although it is critically dependent on an individual's clinical assessment and further studies, early diagnosis might allow DBS to be implemented sooner rather than later, especially in *GBA* mutation carriers.[49,50] Cognitive dysfunction is part of the natural history of PD, and such medical comorbidities increase with age and debility. Hence, a definite genetic diagnosis may enable earlier therapeutic intervention and thus enhance the quality of life for patients.

In this cohort, the most patients remain idiopathic, suggesting that there are additional genetic and/or environmental contributions. Although we have nominated additional sequence and copy number variability, its significance without segregation analysis is unclear. Currently, further affected family members are not available to study. Far larger case and control series are needed to elucidate the relevance of these rare variants to disease or to test the hypothesis that parkinsonism is an oligogenic or multifactorial trait.

In the absence of reliable diagnostic markers, a clinical diagnosis of parkinsonism is based on characteristic features that may be quite subtle early in the disease course. A differential diagnosis is especially challenging without longitudinal follow-up. However, both types of diagnoses can be supported by genetic analysis; because a definite diagnosis informs management strategies, treatment, and quality of life. A genetic yield of 14% in EOPD (< 45 years) should warrant genetic testing as a standard in the clinic. Nevertheless, subsequent meta-analyses of larger patient series are recommended to confirm results and identify novel genes with pathogenic mutations. In this effort, several groups have created mechanisms to connect clinicians and researchers with common interests in specific genes and variants. While we encourage their use (https://genematcher.org/about),

doi:10.1002/mdc3.12501

we have also created a disease-specific Web application to enable researchers' access to the exome data generated in this study (http://annex.can.ubc.ca).

Author Roles

1. Research Project: A. Conception, B. Organization, C. Execution; 2. Statistical Analysis: A. Design, B. Execution, C. Review and Critique; 3. Manuscript Preparation: A. Writing the First Draft, B. Review and Critique.

E.K.G.: 1A, 1B, 1C, 2A, 2B, 2C, 3A, 3B
J.T.: 1B, 1C, 2A, 2B, 2C, 3B
M.M.: 1B, 1C, 3B
S.B.: 1B, 1C, 3B
M.S.P.: 3B
M.J.F.: 1A, 2C, 3B
J.O.A.: 1A, 1C, 3B

Acknowledgements

We are grateful to all individuals who generously participated in this study. Also, we appreciate the technical contributions from Daniel M. Evans and Tara Candido.

Disclosures

Ethical Compliance Statement: We confirm that we have read the Journal's position on issues involved in ethical publication and affirm that this work is consistent with those guidelines.

Funding Sources and Conflict of Interest: This research was supported in part by funds from St. Olav's hospital (E.K.G.), Reberg's Legacy (J.O.A.), and Sør-Trøndelag Parkinsonforening (J.O.A.); the Canada Excellence Research Chairs program (M.J.F.); Leading Edge Endowment Funds provided by the Province of British Columbia (M.J.F.); LifeLabs (M.J.F.) and Genome BC support the Dr. Donald Rix BC Leadership Chair (M.J.F.), and the Peter Cundill Foundation (M.J.F.), and Canadian Institutes of Health Research (J.T.). The Faroese project has been supported by the Faroese Research Council and the Faroese and Danish Parkinson Associations. The authors report no other sources of funding and no conflicts of interest.

Financial Disclosures for the previous 12 months: Matthew J. Farrer serves on the scientific advisory boards of Parkinson's Society Canada, Parkinson's UK, and the Michael J. Fox Foundation. The remaining authors report no sources of funding and no conflicts of interest.

References

1. Stern MB, Lang A, Poewe W. Toward a redefinition of Parkinson's disease. *Mov Disord* 2012;27:54–60.
2. Jankovic J. Levodopa strengths and weaknesses. *Neurology* 2002;58(4 Suppl 1):S19–S32.
3. Chaudhuri KR, Yates L, Martinez-Martin P. The non-motor symptom complex of Parkinson's disease: a comprehensive assessment is essential. *Curr Neurol Neurosci Rep* 2005;5:275–283.
4. Emre M. Dementia associated with Parkinson's disease. *Lancet Neurol* 2003;2:229–237.
5. Langston JW. The Parkinson's complex: parkinsonism is just the tip of the iceberg. *Ann Neurol* 2006;59:591–596.
6. Jankovic J, Rajput AH, McDermott MP, Perl DP. The evolution of diagnosis in early Parkinson disease. Parkinson Study Group. *Arch Neurol* 2000;57:369–372.
7. Gibb WR, Lees AJ. The relevance of the Lewy body to the pathogenesis of idiopathic Parkinson's disease. *J Neurol Neurosurg Psychiatry* 1988;51:745–752.
8. Hughes AJ, Daniel SE, Kilford L, Lees AJ. Accuracy of clinical diagnosis of idiopathic Parkinson's disease: a clinico-pathological study of 100 cases. *J Neurol Neurosurg Psychiatry* 1992;55:181–184.
9. Hughes AJ, Daniel SE, Lees AJ. Improved accuracy of clinical diagnosis of Lewy body Parkinson's disease. *Neurology* 2001;57:1497–1499.
10. Jellinger KA. Morphological substrates of parkinsonism with and without dementia: a retrospective clinico-pathological study. *J Neural Transm Suppl* 2007;91–104.
11. Rajput DR. Accuracy of clinical diagnosis of idiopathic Parkinson's disease. *J Neurol Neurosurg Psychiatry* 1993;56:938–939.
12. Klein C. Genetics in dystonia. *Parkinsonism Relat Disord* 2014;20(Suppl 1):S137–S142.
13. Bonifati V, Rizzu P, van Baren MJ, et al. Mutations in the DJ-1 gene associated with autosomal recessive early-onset parkinsonism. *Science* 2003;299:256–259.
14. Kitada T, Asakawa S, Hattori N, et al. Mutations in the parkin gene cause autosomal recessive juvenile parkinsonism. *Nature* 1998;392:605–608.
15. Valente EM, Abou-Sleiman PM, Caputo V, et al. Hereditary early-onset Parkinson's disease caused by mutations in PINK1. *Science* 2004;304:1158–1160.
16. Edvardson S, Cinnamon Y, Ta-Shma A, et al. A deleterious mutation in DNAJC6 encoding the neuronal-specific clathrin-uncoating co-chaperone auxilin, is associated with juvenile parkinsonism [serial online]. *PLoS ONE* 2012;7:e36458.
17. Paisan-Ruiz C, Bhatia KP, Li A, et al. Characterization of PLA2G6 as a locus for dystonia-parkinsonism. *Ann Neurol* 2009;65:19–23.
18. Ramirez A, Heimbach A, Grundemann J, et al. Hereditary parkinsonism with dementia is caused by mutations in ATP13A2, encoding a lysosomal type 5 P-type ATPase. *Nat Genet* 2006;38:1184–1191.
19. Shojaee S, Sina F, Banihosseini SS, et al. Genome-wide linkage analysis of a Parkinsonian-pyramidal syndrome pedigree by 500 K SNP arrays. *Am J Hum Genet* 2008;82:1375–1384.
20. Trinh J, Farrer M. Advances in the genetics of Parkinson disease. *Nat Rev Neurol* 2013;9:445–454.
21. Hughes AJ, Ben-Shlomo Y, Daniel SE, Lees AJ. What features improve the accuracy of clinical diagnosis in Parkinson's disease: a clinicopathologic study. *Neurology* 1992;42:1142–1146.
22. Aasly JO, Toft M, Fernandez-Mata I, et al. Clinical features of LRRK2-associated Parkinson's disease in central Norway. *Ann Neurol* 2005;57:762–765.
23. Wang K, Li M, Hakonarson H. ANNOVAR: functional annotation of genetic variants from high-throughput sequencing data [serial online]. *Nucleic Acids Res* 2010;38:e164.
24. Kircher M, Witten DM, Jain P, O'Roak BJ, Cooper GM, Shendure J. A general framework for estimating the relative pathogenicity of human genetic variants. *Nat Genet* 2014;46:310–315.
25. Wang K, Li M, Hadley D, et al. PennCNV: an integrated hidden Markov model designed for high-resolution copy number variation detection in whole-genome SNP genotyping data. *Genome Res* 2007;17:1665–1674.
26. Thomas B, Beal MF. Parkinson's disease. *Hum Mol Genet* 2007;16 Spec No. 2:R183–R194.
27. Ylikotila P, Tiirikka T, Moilanen JS, Kaariainen H, Marttila R, Majamaa K. Epidemiology of early-onset Parkinson's disease in Finland. *Parkinsonism Relat Disord* 2015;21:938–942.
28. Trinh J, Amouri R, Duda JE, et al. Comparative study of Parkinson's disease and leucine-rich repeat kinase 2 p. G2019S parkinsonism. *Neurobiol Aging* 2014;35:1125–1131.
29. Trinh J, Farrer M, Ross OA, Guella I. LRRK2-related Parkinson disease In: Pagon RA, Adam MP, Ardinger HH, editors. *GeneReviews* [Internet]. Seattle, WA: University of Washington, Seattle; 2006. Available from: https://www.ncbi.nlm.nih.gov/books/NBK1208.

doi:10.1002/mdc3.12501

30. Johansen KK, Hasselberg K, White LR, Farrer MJ, Aasly JO. Genealogical studies in LRRK2-associated Parkinson's disease in central Norway. *Parkinsonism Relat Disord* 2010;16:527–530.

31. Johansen KK, Jorgensen JV, White LR, Farrer MJ, Aasly JO. Parkinson-related genetics in patients treated with deep brain stimulation. *Acta Neurol Scand* 2011;123:201–206.

32. Angeli A, Mencacci NE, Duran R, et al. Genotype and phenotype in Parkinson's disease: lessons in heterogeneity from deep brain stimulation. *Mov Disord* 2013;28:1370–1375.

33. Davis MY, Johnson CO, Leverenz JB, et al. Association of GBA mutations and the E326K polymorphism with motor and cognitive progression in Parkinson disease. *JAMA Neurol* 2016;73:1217–1224.

34. Alcalay RN, Caccappolo E, Mejia-Santana H, et al. Cognitive performance of GBA mutation carriers with early-onset PD: the CORE-PD study. *Neurology* 2012;78:1434–1440.

35. Tayebi N, Walker J, Stubblefield B, et al. Gaucher disease with parkinsonian manifestations: does glucocerebrosidase deficiency contribute to a vulnerability to parkinsonism? *Mol Genet Metab* 2003;79:104–109.

36. Schneider SA, Paisan-Ruiz C, Garcia-Gorostiaga I, et al. GLUT1 gene mutations cause sporadic paroxysmal exercise-induced dyskinesias. *Mov Disord* 2009;24:1684–1688.

37. Overweg-Plandsoen WC, Groener JE, Wang D, et al. GLUT-1 deficiency without epilepsy–an exceptional case. *J Inherit Metab Dis* 2003;26:559–563.

38. Bawazir WM, Gevers EF, Flatt JF, et al. An infant with pseudohyperkalemia, hemolysis, and seizures: cation-leaky GLUT1-deficiency syndrome due to a SLC2A1 mutation. *J Clin Endocrinol Metab* 2012;97:E987–E993.

39. Pons R, Collins A, Rotstein M, Engelstad K, De Vivo DC. The spectrum of movement disorders in Glut-1 deficiency. *Mov Disord* 2010;25:275–281.

40. Wang D, Pascual JM, De Vivo D. Glucose transporter type 1 deficiency syndrome. In: Pagon RA, Adam MP, Ardinger HH, et al. , editors. *GeneReviews* [Internet]. Seattle, WA: University of Washington, Seattle; 2002. Available from: https://www.ncbi.nlm.nih.gov/books/NBK1430/.

41. Klepper J, Leiendecker B. Glut1 deficiency syndrome and novel ketogenic diets. *J Child Neurol* 2013;28:1045–1048.

42. Kass HR, Winesett SP, Bessone SK, Turner Z, Kossoff EH. Use of dietary therapies amongst patients with GLUT1 deficiency syndrome. *Seizure* 2016;35:83–87.

43. Alter AS, Engelstad K, Hinton VJ, et al. Long-term clinical course of Glut1 deficiency syndrome. *J Child Neurol* 2015;30:160–169.

44. Leen WG, Mewasingh L, Verbeek MM, Kamsteeg EJ, van de Warrenburg BP, Willemsen MA. Movement disorders in GLUT1 deficiency syndrome respond to the modified Atkins diet. *Mov Disord* 2013;28:1439–1442.

45. Ross JP, Dupre N, Dauvilliers Y, et al. Analysis of DNAJC13 mutations in French-Canadian/French cohort of Parkinson's disease. *Neurobiol Aging* 2016;45:212.e13–212.e17.

46. Gustavsson EK, Trinh J, Guella I, et al. DNAJC13 genetic variants in parkinsonism. *Mov Disord* 2015;30:273–278.

47. Rubio JP, Topp S, Warren L, et al. Deep sequencing of the LRRK2 gene in 14,002 individuals reveals evidence of purifying selection and independent origin of the p.Arg1628Pro mutation in Europe. *Hum Mutat* 2012;33:1087–1098.

48. Klein C, Marras C, Munchau A. Dystonia overview. In: Pagon RA, Adam MP, Ardinger HH, et al. , editors. *GeneReviews* [Internet]. Seattle, WA: University of Washington, Seattle; 2014. Available from: https://www.ncbi.nlm.nih.gov/books/NBK1155.

49. Moro E, Allert N, Eleopra R, et al. A decision tool to support appropriate referral for deep brain stimulation in Parkinson's disease. *J Neurol* 2009;256:83–88.

50. Wachter T, Minguez-Castellanos A, Valldeoriola F, Herzog J, Stoevelaar H. A tool to improve pre-selection for deep brain stimulation in patients with Parkinson's disease. *J Neurol* 2011;258:641–646.

Supporting Information

Additional Supporting Information may be found online in the supporting information tab for this article:

Figure S1. Map showing the region of central Norway, including Nord-Trøndelag and Sør-Trøndelag, from which 108 patients with early onset Parkinson's disease were recruited.

Figure S2. Venn diagram of family history of parkinsonism among the 108 patients with early onset parkinsonism from Norway.

doi:10.1002/mdc3.12501