The clinical and genetic heterogeneity of paroxysmal dyskinesias

Alice R. Gardiner,1,2 Fatima Jaffer,1,2 Russell C. Dale,3 Robyn Labrum,4 Roberto Erro,5 Esther Meyer,6 Georgia Xiromerisiou,2,7 Maria Stamelou,5,8,9 Matthew Walker,10 Dimitri Kullmann,10 Tom Warner,2 Paul Jarman,5 Mike Hanna,1 Manju A. Kurian,6,11 Kailash P. Bhatia5,* and Henry Houlden1,2,4,*

These authors contributed equally to this work.

Paroxysmal dyskinesia can be subdivided into three clinical syndromes: paroxysmal kinesigenic dyskinesia or choreoathetosis, paroxysmal exercise-induced dyskinesia, and paroxysmal non-kinesigenic dyskinesia. Each subtype is associated with the known causative genes PRRT2, SLC2A1 and PNKD, respectively. Although separate screening studies have been carried out on each of the paroxysmal dyskinesia genes, to date there has been no large study across all genes in these disorders and little is known about the pathogenic mechanisms. We analysed all three genes (the whole coding regions of SLC2A1 and PRRT2 and exons one and two of PNKD) in a series of 145 families with paroxysmal dyskinesias as well as in a series of 53 patients with familial episodic ataxia and hemiplegic migraine to investigate the mutation frequency and type and the genetic and phenotypic spectrum. We examined the mRNA expression in brain regions to investigate how selective vulnerability could help explain the phenotypes and analysed the effect of mutations on patient-derived mRNA. Mutations in the PRRT2, SLC2A1 and PNKD genes were identified in 72 families in the entire study. In patients with paroxysmal movement disorders 68 families had mutations (47%) out of 145 patients. PRRT2 mutations were identified in 35% of patients, SLC2A1 mutations in 10%, PNKD in 2%. Two PRRT2 mutations were in familial hemiplegic migraine or episodic ataxia and hemiplegic migraine to investigate the mutation frequency and type and the genetic and phenotypic spectrum. We examined the mRNA expression in brain regions to investigate how selective vulnerability could help explain the phenotypes and analysed the effect of mutations on patient-derived mRNA. Mutations in the PRRT2, SLC2A1 and PNKD genes were identified in 72 families in the entire study. In patients with paroxysmal movement disorders 68 families had mutations (47%) out of 145 patients. PRRT2 mutations were identified in 35% of patients, SLC2A1 mutations in 10%, PNKD in 2%. Two PRRT2 mutations were in familial hemiplegic migraine or episodic ataxia and hemiplegic migraine alone. Several previously unreported mutations were identified. The phenotypes associated with PRRT2 mutations included a high frequency of migraine and hemiplegic migraine. SLC2A1 mutations were associated with variable phenotypes including paroxysmal kinesigenic dyskinesia, paroxysmal non-kinesigenic dyskinesia, episodic ataxia and myotonia and we identified a novel PNKD gene deletion in familial hemiplegic migraine. We found that some PRRT2 loss-of-function mutations cause nonsense mediated decay, except when in the last exon, whereas missense mutations do not affect mRNA. In the PNKD family with a novel deletion, mRNA was truncated losing the C-terminus of PNKD-L and still likely loss-of-function, leading to a reduction of the inhibition of exocytosis, and similar to PRRT2, an increase in vesicle release. This study highlights the frequency, novel mutations and clinical and molecular spectrum of PRRT2, SLC2A1 and PNKD mutations as well as the phenotype-genotype overlap among these paroxysmal movement disorders. The investigation of paroxysmal movement disorders should always include the analysis of all three genes, but around half of our paroxysmal series remain genetically undefined implying that additional genes are yet to be identified.

1 MRC Centre for Neuromuscular Diseases, UCL Institute of Neurology, Queen Square, London WC1N 3BG, UK
2 Department of Molecular Neuroscience, UCL Institute of Neurology, Queen Square, London WC1N 3BG, UK
3 Paediatrics and Child Health, Children’s Hospital, Westmead, University of Sydney, Australia
4 Neurogenetics Laboratory, UCL Institute of Neurology, Queen Square, London WC1N 3BG, UK
5 Department of Motor Neuroscience and Movement Disorders, UCL Institute of Neurology, Queen Square, London WC1N 3BG, UK
6 Developmental Neurosciences, UCL Institute of Child Health, London WC1N 3JH, UK
7 Department of Neurology, Papageorgiou Hospital, Thessaloniki University of Athens, Greece

Received May 22, 2015. Revised August 12, 2015. Accepted August 27, 2015
© The Author (2015). Published by Oxford University Press on behalf of the Guarantors of Brain.
This is an Open Access article distributed under the terms of the Creative Commons Attribution License (http://creativecommons.org/licenses/by/4.0/), which permits unrestricted reuse, distribution, and reproduction in any medium, provided the original work is properly cited.
Introduction

Paroxysmal dyskinesia was first reported in 1892 by Shuzo Kure in a 23-year-old Japanese man, who had frequent movement-induced paroxysmal attacks from the age of 10 years. At that time the diagnosis was referred to as atypical Thomsen’s disease (Kure, 1892). Later, Gowers (1901) described a similar child, but he considered this movement disorder an epileptic phenomenon, and in 1940, Mount and Reback (1940) described a 23-year-old with involuntary writhing and posturing of the trunk and extremities and labelled this condition paroxysmal dystonic choreoathetosis. Kertesz (1967) and Weber (1967) described families with this condition termed paroxysmal kinesigenic choreo-athetosis and familial paroxysmal dystonia, and Demirkiran and Jankovic (1995) amalgamated the many terms used, suggesting three subtypes, comprising paroxysmal kinesigenic (PKD or PKC), non-kinesigenic (PNKD), and exercise-induced dyskinesia (PED) (Bruno et al., 2004, 2007; Bhatia, 2011). A fourth type, paroxysmal hypnogenic dyskinesia (PHD), characterized by attacks of dyskinesia during sleep, was previously included, but has since been recognized as autosomal dominant nocturnal frontal lobe epilepsy (Sohn and Lee, 2011).

The most common of the paroxysmal movement disorders is PKD, in which attacks are precipitated by voluntary movements such as standing from a sitting position, or the transition from walking to running. Onset is usually in childhood, and attacks are often controlled by carbamazepine (Bhatia, 2001, 2011; Erro et al., 2014). PKD is frequently preceded by infantile convulsions, often with choreoathetosis. The gene responsible for PKD proved elusive for many years, but was recently identified as PRRT2, which encodes a small proline-rich transmembrane protein (Chen et al., 2011; Wang et al., 2011; Cloarec et al., 2012; de Vries et al., 2012; Gardiner et al., 2012; Guerrini and Mink, 2012; Hedera et al., 2012; Heron et al., 2012; Li et al., 2012; Liu et al., 2012; Scheffer et al., 2012). The function of the protein is unknown, but it has been shown to interact with the synaptic protein SNAP25 (Lee et al., 2012). Mutations in the PRRT2 gene account for a large proportion of PKD and several groups have reported mutations in this gene (Chen et al., 2011; Wang et al., 2011; Cao et al., 2012; de Vries et al., 2012; Friedman et al., 2012; Gardiner et al., 2012; Heron et al., 2012; Lee et al., 2012; Li et al., 2012; Liu et al., 2012; Ono et al., 2012; Ishii et al., 2013; Specchio et al., 2013).

Attacks of PNKD are usually triggered by alcohol, coffee or strong emotion. They last longer than attacks of PKD, often from 10 min to 1 h, but can last as long as 12 h. However, they are much more infrequent and occur only a few times a year (Mount and Reback, 1940; Bhatia, 1999; Lombroso and Fischman, 1999; Vercueil, 2000; Lee et al., 2004; Engelen and Tijsse, 2005; Friedman et al., 2009; Ghezzi et al., 2009; van Rootselaar et al., 2009; Benz et al., 2012; Pons et al., 2012). The gene responsible for PNKD was identified as the MR-1 gene in 2004, but it is now referred to as PNKD (Raskind et al., 1998; Lee et al., 2004; Rainier et al., 2004). To date three mutations in this gene have been reported: p.A7V, p.A9V and p.A33P, the first two of which have been found in multiple unrelated patients (Lee et al., 2004; Friedman et al., 2009; Ghezzi et al., 2009; Shen et al., 2011; Pons et al., 2012; Erro et al., 2014). Recent work from Shen et al., (2015) has shown that PNKD interacts with the synaptic active zone proteins RAB-interacting molecule (RIM)1 and RIM2, and modulates neurotransmitter release. The mutant protein is less effective at inhibiting exocytosis.

Lance (1977) described a family with exercise-induced dystonia with attacks lasting between 5 and 30 min, once or twice per month. This disorder is now termed PED (Lance, 1977). PED is thought to be the rarest of the three paroxysmal movement disorders, where attacks are induced by physical exertion after long periods of exercise. The condition can be associated with migraine, hemiplegia, ataxia and epilepsy (Zorzi et al., 2003; Bhatia, 2011). Mutations in the SLC2A1 gene, which encodes the glucose transporter type 1 protein, have recently been found to be responsible for causing PED, often called GLUT1 deficiency.
syndrome 2 (Wang et al., 2000; Vermeer et al., 2007; Suls et al., 2008). SLC2A1 mutations also cause GLUT1 deficiency syndrome 1, a phenotypically variable syndrome that often includes ataxia, microcephaly, intellectual dysfunction, dystonia, epilepsy and low fasting glucose levels detected on CSF analysis (Wang et al., 2000; Vermeer et al., 2007; Suls et al., 2008; Schneider et al., 2009; Fung et al., 2011; Gokben et al., 2011; Hashimoto et al., 2011; Bawazir et al., 2012; Agostinelli et al., 2013; Muhle et al., 2013; Weller et al., 2015).

The majority of published reports on paroxysmal movement disorders are single families, small series or single gene studies with little known about the gene mechanisms. Here, we carry out the first large screening study of the three main paroxysmal dyskinesia genes [the total coding regions of SLC2A1 and PRRT2 and exons one and two (the only exons in which mutations have been previously identified) of PNKD] in a large referral series of 145 paroxysmal movement disorders and in a further 53 genetically undefined patients with episodic ataxia or familial hemiplegic migraine. We identify the mutation frequency and spectrum as well as genetic and phenotypic heterogeneity, describe novel mutations, and investigate the mutation mechanisms amongst the paroxysmal dyskinesias.

Materials and methods

Patients and unaffected family members were recruited through the laboratory with consent and ethical approval (NHNN studies 06/N076 and 07/Q0512/26); they were seen either at the National Hospital in Queen Square, or referred from other centres for genetic testing with local approval. Patients were diagnosed with a paroxysmal dyskinesia or movement disorder based on recognized criteria (Bruno et al., 2004, 2007; Kinali et al., 2004; Bhatia, 2011) by the authors. Acquired causes were excluded using clinical investigation prior to genetic testing. Episodic ataxia and familial hemiplegic migraine cases were negative for mutations in the KCNA1 and CACNA1A genes by direct sequencing of all codons. DNA was extracted from blood of affected patients and unaffected family members using standard diagnostic laboratory methods.

Sequencing

Polymerase chain reaction (PCR) was used to amplify the three coding exons and flanking introns of the PRRT2 gene, the 10 coding exons and flanking introns of the SLC2A1 gene, and the first two coding exons and flanking introns of the PNKD gene (Supplementary Table 1). For each gene the longest transcript was used for primer design and sequencing: PRRT2-001: ENST00000358758; SLC2A1-001: ENST00000426263; PNKD-001: ENST00000273077. PCR amplification was performed using 10 pmol of both forward and reverse genomic primers (synthesized by Sigma-Aldrich) and FastStart™ Taq DNA polymerase (Roche). Each purified product was then sequenced using forward or reverse primers, as well as internal sequencing primers to ensure complete coverage of the case of exon 2 of PRRT2 with Applied Biosystems BigDye® terminator v3.3 sequencing chemistry as per the manufacturer’s instructions. The resulting reactions were resolved on an ABI3730XL genetic analyser (Applied Biosystems) and analysed with SeqScape v2.5 software (Gene codes).

In developing our genetic analysis strategy for diagnostics we also developed a custom Illumina sequencing gene panel (Illumina Inc.). This panel included the PRRT2, SLC2A1 and PNKD genes. These genes had a mean coverage of 269 ×, 196 × and 178 ×, respectively and 24 samples were analysed in this way. All regions of the genes were covered and no coverage gaps had to be completed by Sanger sequencing. The analysis of data consisted of mapping the raw data to the hg19 human reference assembly using Novoalign software, and PCR duplicates were removed using the Picard software. Indels were called using the GATK package and variants annotated using SAMtools. Mutations were verified in both directions. Mutation position was labelled from the transcriptional start site of the genes, according to the standard nomenclature.

Expression methods

Regional distribution of PRRT2, SLC2A1, PNKD, KCN1A, SNAP25 and CACNA1A mRNA expression in the normal human brain was determined using microarray analysis of human post-mortem brain tissue from the UK Human Brain Expression Consortium (Trabzuni et al., 2011). Brain tissues originating from 134 control Caucasian individuals were collected by the Medical Research Council (MRC) Sudden Death Brain and Tissue Bank (Edinburgh, UK). The following brain regions were included in the analysis: cerebellum, frontal cortex, hippocampus, medulla, occipital cortex, putamen, substantia nigra, temporal cortex, thalamus and white matter. Total RNA was isolated from these tissues using mRNeasy 96-well kit (Qiagen) before processing with the Ambion® WT Expression Kit and Affymetrix GeneChip Whole Transcript Sense Target Labeling Assay, and hybridization to the Affymetrix Exon 1.0 ST Array. The probe set defining each gene mRNA was determined using the Affymetrix NetAffx annotation file (HuEx-1.0-st-v2 Probe set Annotations, Release 31). The combined signal of the gene probe sets were used to determine mRNA expression.

Sequencing of PNKD and PRRT2 cDNA from affected patient fibroblast mRNA was carried out to assess the presence of nonsense-mediated decay and to indicate the presence of a truncated protein in mutations that affect the last exon of the gene. Fibroblasts were first taken with informed consent and mRNA was extracted using a Qiagen miRNA kit. cDNA was synthesized from affected patients and unaffected family members using standard diagnostic laboratory methods.

Results

Mutations in the PRRT2 gene were found in 53 families or sporadic cases, with nine different mutation types (Figs 1–5
Figure 1  Genetic structure and mutations in PRRT2, SLC2A1 and PNKD. Schematic diagrams of the PRRT2 (A), SLC2A1 (B) and PNKD (C) genes. In each case mutations that have been previously reported to cause a paroxysmal movement disorder are shown above the gene, and mutations found in this paper are shown below (blue have previously been reported, red are novel).

Figure 2  Family tree and mutation chromatograms. Filled symbols indicate family members that are affected, unfilled symbols are unaffected. The proband is indicated with a black arrow. +/− denotes an individual that is heterozygous for the mutation shown, −/− does not carry the mutation.
A male to female ratio of 2:1.3 was identified in those expressing a phenotype, and the patient demographic was 56% British and a mixture of other populations accounting for the other 44%. As widely reported, by far the most common mutation (44 families, 82%) was an insertion of a cytosine into a string of nine cytosines, resulting in a frame shift mutation and premature stop codon (p.R217Pfs*8). Each of the other nine mutations accounted for one family and the majority were loss-of-function. These mutations were found in families with a number of different ethnicities and there was no common background haplotype. Four mutations were novel and two of the mutations (p.G305W and p.C332_V333insD) have only been reported by us in the past. We include them here, as well as the cases with p.R217Pfs*8 mutations, for the assessment of the frequency of PRRT2 mutations in our cohort (Gardiner et al., 2012; Silveira-Moriyama et al., 2013). The p.P215R variant is also included in the mutation table; it has a frequency of <7:10 000 in the ExAC database and not seen in 488 UK control subjects. The pathogenicity of this change is still uncertain. The p.P216H variant has been found in our patient series but was also found in the UK control population at a rate of 1%. Mutations in the PRRT2 gene were mainly associated with paroxysmal kinesigenic dyskinesia with a number of associated phenotypes (Table 1) including: (i) episodic ataxia; (ii) benign epilepsy; (iii) PED; and (iv) migraine and familial hemiplegic migraine. Fifty-one patients were part of the paroxysmal dyskinesia series and the remaining two were from the episodic ataxia and familial hemiplegic migraine series.

Migraine and hemiplegic migraine were by far the most common associated phenotypes (Table 1). Interestingly, the majority of patients were given symptomatic treatment, mainly with carbamazepine; it has been widely reported that patients with PRRT2-positive PKD are more likely to respond well to the drug than patients without a mutation (Li et al., 2013; Mao et al., 2014). There did not appear to be a correlation between genotype and efficacy of treatment in our cohort. Initially the extended Indian families were taking phenytoin, which was then usually switched to carbamazepine, and lamotrigine in one patient. Depending on availability some of the extended Indian family patients still take phenytoin. Patient 48, who did appear to benefit from even high doses of the drugs. No treatment was being given in three families, at patients’ request. A family with episodic ataxia and one with familial hemiplegic migraine alone were identified with PRRT2 mutations. The familial hemiplegic migraine family proband presented as an infant with infrequent seizures until age 2 years and then developed...
Figure 4 Mutation effect in PRRT2 and PNKD frameshift mutations. (A) Schematic diagram of PRRT2 showing the elongation of the protein caused by p.*341Lext27, and the chromatogram identifying the mutation in the patient DNA with no NMD in mRNA from this family. (B) Schematic diagram of the wild-type and truncated PNKD-L, the result of the p.P341Pfs*2 mutation. The cDNA sequencing (B) shows the mutation was present at the mRNA level (top = forward sequencing, bottom = reverse sequencing in the lower figure) and so excludes the possibility of nonsense-mediated decay.

Figure 5 Likely mechanism of action of paroxysmal dyskinesia genes. A suggested mechanism for the paroxysmal dyskinesia genes, where mutations in PRRT2, PNKD and SLC2A1 result in disruption of neurotransmitter release regulation and thus impaired synaptic release. Circles indicate presynaptic vesicles containing neurotransmitter (dots). Yellow vesicles are affected by SLC2A1 mutations, green by PNKD mutations and blue by PRRT2 mutations.
| Patient | Ethnicity | Age at onset/ current age | Affected cases and gender | Phenotype in the proband | Family history | Family members tested for segregation | Genetics | Frequency in ExAC | Previously reported (reference) |
|---------|-----------|---------------------------|----------------------------|---------------------------|----------------|--------------------------------------|----------|------------------|---------------------------------|
| 1       | Somalia   | 12–13/24–27               | 1M 1F                      | PKD with seizures         | Affected sister | Yes                                  | p.L171Lfs*3 | 0                | Chen et al., 2011                |
| 2       | British   | 7–8/12–16                 | 1M 1F                      | PKD, one unaffected with the mutation, | Yes, affected sister, mother unaffected carrier | Yes                                  | p.R217X     | 0                | Liu et al., 2012                |
| 3       | Austrian  | 0.5–27/29–51              | 1M 1F                      | PKD                        | Yes, affected sister, father unaffected carrier | Yes                                  | p.R217Pfs*8 | 0.006            | Chen et al., 2011; Wang et al., 2011; Lee et al., 2012 |
| 4       | Wales/ India | 6–11/18–49             | 4M                          | PKD, Migraine with aura    | Yes, affected paternal grandfather, father, brother with migraine | No                        | p.R217Pfs*8 | 0.006            | As above for p.R217Pfs*8         |
| 5       | Ireland   | 8/42–45                   | 1M 1F                      | PKD                        | Affected sister | Yes                                  | p.R217Pfs*8 | 0.006            | Chen et al., 2011                |
| 6       | British   | 1–6/12–62                 | 1M 1F                      | PKD, several individuals with HM and classical migraine | Yes, autosomal dominant family | Yes                                  | p.R217Pfs*8 | 0.006            | Wang et al., 2011; Lee et al., 2012 |
| 7       | British   | 0.5–8/12–52               | 2M 2F                      | Benign familial infantile epilepsy, HM | Yes, father, sister and cousins affected with HM | No                        | p.R217Pfs*8 | 0.006            |                                      |
| 8       | Pakistan  | 14/31–33                  | 1M 1F                      | PKD                        | Yes, affected sister | No                                  | p.R217Pfs*8 | 0.006            |                                      |
| 9       | British   | 4–10/12–56                | 2M 2F                      | PKD, meningois and recurrent seizures as a child | Yes, brother and sister possibly affected, affected mother | Yes                                  | p.R217Pfs*8 | 0.006            |                                      |
| 10      | British   | 6–11/20–68                | 2M 2F                      | PKD with migraine          | Autosomal dominant family history. Seizures in one case. | Yes                                  | p.R217Pfs*8 | 0.006            |                                      |
| 11      | British   | 6–16/8–38                 | 1M 1F                      | PKD                        | Probable, mother migraine | No                                  | p.R217Pfs*8 | 0.006            |                                      |
| 12      | Turkey    | 5/16                      | 1M                          | PKD                        | No family history | No                                  | p.R217Pfs*8 | 0.006            |                                      |
| 13      | British   | 12/18                     | 1F                          | PKD                        | No family carrier | No                                  | p.R217Pfs*8 | 0.006            |                                      |
| 14      | British   | 10–12/59                  | 1M 2F                      | Episodic ataxia with familial hemiplegic migraine | Yes, affected mother and children with familial hemiplegic migraine | No                        | p.R217Pfs*8 | 0.006            |                                      |
| 15      | Pakistan  | 8/40–42                   | 2M                          | PKD, both brothers have migraine with aura | Yes, affected brother | No                                  | p.R217Pfs*8 | 0.006            |                                      |
| 16      | Malta     | 8–18/25–48                | 1M 1F                      | PKD with migraine          | Yes, affected mother | Yes                                  | p.R217Pfs*8 | 0.006            |                                      |
| 17      | Pakistan  | 8/43                      | 2M                          | PKD with headaches         | Yes, affected twin brother | No                                  | p.R217Pfs*8 | 0.006            |                                      |
| 18      | British   | 27/48                     | 1M                          | PKD                        | No                          | No                                  | p.R217Pfs*8 | 0.006            |                                      |
| 19      | Singapore | 9–12/42–47                | 1M 1F                      | PKD                        | Yes, daughter has childhood seizures | No                        | p.R217Pfs*8 | 0.006            |                                      |
| 20      | India     | 6–14/12–42                | 3M                          | PKD with migraine          | Yes, affected brother and father, family history of seizures | Yes                                  | p.R217Pfs*8 | 0.006            |                                      |
| 21      | British   | 0.5–30/87                 | 3M 3F                      | PKD, Migraine, HM, epilepsy. Three mutation carriers asymptomatic | Yes, dominant, large number affected | Yes                                  | p.R217Pfs*8 | 0.006            |                                      |
| 22      | British   | 12–18/14–39               | 1M 1F                      | PKD                        | Yes, mother has migraine | No                                  | p.R217Pfs*8 | 0.006            |                                      |
| 23      | British   | 14/39                     | 1M                          | PKD                        | No                          | No                                  | p.R217Pfs*8 | 0.006            |                                      |
| 24      | India     | 7–13/9–60                 | 9M 6F                      | PKD with seizures in many as a child. Several mutation carriers are asymptomatic | Yes, large autosomal dominant family history | Yes                                  | p.R217Pfs*8 | 0.006            |                                      |
| 25      | India     | 8/12–52                   | 2M 1F                      | PKD with migraine          | Yes, father affected and seizures in paternal aunt | Yes                                  | p.R217Pfs*8 | 0.006            |                                      |
| 26      | British   | 8–12/28–76                | 5M 8F                      | PKD with hemiplegic migraine and seizures | Yes, large autosomal dominant family history | Yes                                  | p.R217Pfs*8 | 0.006            |                                      |
| 27      | Slovakia  | 4–7/9–12                  | 2F                          | PKD with migraine and burning hemiplegia | Yes, sister has migraine | No                                  | p.R217Pfs*8 | 0.006            |                                      |
| 28      | British   | 6–8/12–49                 | 1M 2F                      | PKD                        | Yes, two affected relatives | No                                  | p.R217Pfs*8 | 0.006            |                                      |
| 29      | British   | 9/32                      | 1F                          | PKD                        | Yes, mother with migraine, uncle with infantile convulsion | No                                  | p.R217Pfs*8 | 0.006            |                                      |
| 30      | British   | 9/19                      | 1F                          | PKD                        | No                          | No                                  | p.R217Pfs*8 | 0.006            |                                      |
| Patient | Ethnicity  | Age at onset/ | Affected | Affected | Phenotype in the proband | Family history | Family members tested for segregation | Genetics | Frequency in ExAC | Previously reported (reference) |
|---------|------------|---------------|----------|----------|---------------------------|----------------|-------------------------------------|----------|-------------------|----------------------------------|
| 31      | British    | 11/20         | 1F       | PKD      | Mother is carrier; she had single episode of torticollis but no paroxysmal movement disorder | Yes            | p.R217Pfs*8                   | 0.006    |                   |                                   |
| 32      | Pakistani  | 10/18–49      | 1M       | PKD      | Yes, affected father with PKD | Yes            | p.R217Pfs*8                   | 0.006    |                   |                                   |
| 33      | Irish      | 6–12/31–59    | 1M 1F    | Infantile convulsions with HM | Yes, Multiple affected members with PKD, infantile convulsions and/or HM | Yes            | p.R217Pfs*8                   | 0.006    |                   |                                   |
| 34      | Irish      | 0.5–5/8–40    | 1F 2M    | ICCA later PKD and migraine | Yes, affected brother, father mutation carrier but no history of ICCA | Yes            | p.R217Pfs*8                   | 0.006    |                   |                                   |
| 35      | British    | 0.5/2         | 1M       | Infantile seizures | Yes, affected father with PKD | No             | p.R217Pfs*8                   | 0.006    |                   |                                   |
| 36      | British    | 39/63         | 1M       | PKD and episodic ataxia with dysarthria | No, No         | p.R217Pfs*8                   | 0.006    |                   |                                   |
| 37      | British    | 3/8           | 1M       | PKD      | No                           | No             | p.R217Pfs*8                   | 0.006    |                   |                                   |
| 38      | British    | 12/29         | 1M       | PKD      | No                           | No             | p.R217Pfs*8                   | 0.006    |                   |                                   |
| 39      | Sri Lanka  | 6/16          | 1M       | PKD      | No                           | No             | p.R217Pfs*8                   | 0.006    |                   |                                   |
| 40      | Afghanistan| 8/15          | 1M       | PKD and HM | No                           | No             | p.R217Pfs*8                   | 0.006    |                   |                                   |
| 41      | Pakistan   | 8/27          | 1M       | PKD      | N/A                          | No             | p.R217Pfs*8                   | 0.006    |                   |                                   |
| 42      | Australia  | 5/10          | 2M       | PKD and hemiplegic migraine | Yes, father had hemiplegic migraine | No             | p.R217Pfs*8                   | 0.006    |                   |                                   |
| 43      | British    | 7–14/22–49    | 1M 1F    | PKD      | Yes, mother                  | Yes            | p.R217Pfs*8                   | 0.006    |                   |                                   |
| 44      | British    | 14/33         | 1M       | PKC or PKD | No                           | No             | p.R217Pfs*8                   | 0.006    |                   |                                   |
| 45      | British    | 7–12/9–32     | 2F       | PKC and hemiplegic migraine | Yes mother       | No             | p.R217Pfs*8                   | 0.006    |                   |                                   |
| 46      | British    | 8–14/12–37    | 1M 1F    | PKC      | Yes mother                   | No             | p.R217Pfs*8                   | 0.006    |                   |                                   |
| 47      | British    | 11/21         | 1M       | PKD      | No                           | No             | R240X                        | 0        |                   | Cloarec et al., 2012; Lee et al., 2012. |
| 48      | British    | NA/23         | 1F       | PKD      | No                           | Yes            | p.G305W                       | 0        |                   | Liu et al., 2012                   |
| 49      | India      | 14/46         | 2M       | PKD and migraine | Yes, father had seizures as a child | No             | c.997_998insATG; p.C332_V333insD | 0.00002  |                   | Gardiner et al., 2012             |
| 50      | British    | 2/15          | 1F       | HM and benign seizures      | Yes, several   | No             | c.1011C > T (exon 3 splice site) | No       |                   | Liu et al., 2012                   |
| 51      | British    | 12–18/16–35   | 2F 2M    | PKD, migraine with aura (visual and hemisensory) | Yes, family history of migraine with aura and epilepsy | Yes            | p.*341Lext27                   | 0        |                   | Gardiner et al., 2012             |
| 52      | British    | 15/32         | 1F       | PKD, migraine aura | No                  | No             | p.P215R                       | 0.0008   |                   | Gardiner et al., 2012             |
| 53      | Mauritius  | 8–24/16–62    | 2F 6M    | PKD and migraine | Yes multiple | Yes            | p.P215R                       | 0.0008   |                   | Gardiner et al., 2012             |

HM = hemiplegic migraine; ICCA = infantile convulsions and paroxysmal choreoathetosis. Variants were seen that cause amino acid changes E23K but this was non-pathogenic as seen in controls. P215H and R216H are of unknown pathogenicity as seen in the control population and R216H was in a patient with a definite SLC2A1.
typical hemiplegic migraine attacks. The sister, father and two cousins also had classical hemiplegic migraine and the attacks in the proband persisted until now (aged 18 years) but responded to carbamazepine.

Fourteen SLC2A1 mutations were identified in the paroxysmal dyskinesia series (10%) and one in the episodic ataxia and familial hemiplegic migraine series (Figs 1, 2 and Table 2). In general these were complex cases that had been heavily investigated prior to obtaining a genetic diagnosis. Eight had PED, often associated with other features such as epilepsy and migraine. Three had PKD (one with epilepsy) and one had PNKD, two with episodic ataxia and one with myotonia and dystonia, as discussed below. Eleven of the mutations had previously been reported as being pathogenic. The p.C201R mutation has not before been identified but presented with a PNKD phenotype and was present in the affected mother. p.C201R is not present in population databases, but is not well conserved and predicted to be benign by PolyPhen-2 but damaging by SIFT. p.T60M is present in 0.00015% of the population, is moderately conserved and is predicted to be damaging by PolyPhen-2 but tolerated by SIFT. This mutation has been reported in association with seizures in the past but like in our family, there was reduced penetrance. Patient 66 had sequence variants in both SLC2A1 and PRRT2 (p.R333Q and p.P216H, respectively) and a PKD phenotype, but the PRRT2 mutation is unlikely to be pathogenic as it is present in 1% of controls we analysed, and the p.R333Q mutation has been reported previously as pathogenic.

The majority of PRRT2 mutations are predicted to be loss-of-function and likely lead to haploinsufficiency. It has been previously demonstrated that mutations p.Q163X, p.G192WfsX8 and p.R217PfsX8 result in nonsense-mediated decay (Wu et al., 2014). This is not the case for all mutations as cDNA created from two of our mutations; p.G305W and p.*341Lext27 (a stop codon mutation extending the protein, HGVS standard nomenclature used; den Dunnen and Antonarakis, 2000) do not affect mRNA or lead to a longer transcript (Figs 3 and 4A). The mechanism behind these two mutations is likely to be the same as those causing nonsense-mediated decay with lack of association in the SNAP25/SNARE complex and greater vesicle release (Fig. 5). SLC2A1 mutations were associated with a wide spectrum of clinical features. Family 56 was identified with a novel heterozygous mutation at p.G76V that was not present in 488 controls and 6502 exomes in the exome variant server. This patient was a 26-year-old, diagnosed with attention-deficit hyperactivity disorder as a child and since then has had episodes of ‘wobbly’ eyes, legs and arms, and abnormal arm posturing that last 5–10 min, several times per day. Triggers for these episodes included tiredness, sudden movement, intercurrent infection or illness and excitement. He experienced episodes of weakness and painful cramps in his hands and his legs. He has tried carbamazepine, which helped a little, and acetazolamide may have helped reduce the frequency of these attacks. He underwent repeat long exercise testing (McManis) and this showed significant decrement, accompanied by weakness of the exercised hand muscles. This was most unexpected given that SLC2A1 is best known as a brain transporter; however, there is some evidence of the protein having an additional important role in skeletal muscle (Andrisse et al., 2014). This result was repeated and abnormal spanning over several years. The significant decrement on McManis testing ranged from 51–66%. The clinical diagnosis at that time suggested a periodic paralysis phenotype but the movement disorder was not consistent with this.

In the PNKD gene, four mutations were identified (Figs 1–3 and Table 3). Three were in the paroxysmal dyskinesia series and one in a familial hemiplegic migraine family. The mutations associated with paroxysmal dyskinesias were in phenotypically typical PNKD families with non-kinesigenic precipitants such as stress or strong coffee. These mutations have been reported in the past and these were in two unrelated families with p.A7V and one with p.A9V. In the familial hemiplegic migraine family the mutation was novel and the female proband presented at 42 years of age with a typical attack of hemiplegic migraine with headache, abnormal vision and left-sided motor and sensory weakness that lasted for 45 min to an hour in duration. She had a normal MRI shortly after the event and other cardiac investigations were unremarkable, and the hemiplegic migraine resolved. A few months later she had a similar hemiplegic migraine attack. Her paternal great uncle and father had similar attacks. Her father presented at a similar age and to date has had over 50 hemiplegic migraine attacks, often without a headache. He has presented to the emergency department many times concerned that this was a stroke and has been extensively worked up but imaging and other investigations have been normal. A heterozygous mutation of c.1022delC; p.P341fs*2 was identified in the PNKD gene in the proband and father, not in the mother. We analysed cDNA, from mRNA extracted from patient fibroblasts. The deletion was present in the mRNA, indicating that nonsense-mediated decay would not occur, although nonsense-mediated decay is dependent on cell type and therefore it is possible that it could occur in neurons. This mutation therefore caused the formation of a truncated PNKD in the mRNA (Figs 3 and 4B). Although functional work was not carried out the truncating effect of this mutation is likely to have an abnormal effect on exocytosis due to impaired interaction between PNKD and RIM1 (Fig. 5).

**Discussion**

High prevalences of PRRT2, SLC2A1 and PNKD mutations were identified in this large, mainly London based paroxysmal movement disorder referral series. Although we have a multi-ethnic population the results corroborate smaller individual gene series (Fig. 1 and Tables 1–3).
Table 2  Clinical phenotype and demographics of families and patients with SLC2A1 mutations

| Patient | Ethnicity | Age at onset/ current age | Affected cases and gender | Phenotypic description | Family history | Family members tested for segregation | CSF glucose: blood ratio | Genetics | Frequency in ExAC | Previously reported (reference) |
|---------|-----------|---------------------------|---------------------------|-----------------------|----------------|--------------------------------------|------------------------|----------|----------------|----------------------------------|
| 54      | British   | 5/40                      | IF                        | Exercise induced dystonia, seizures and hemiplegic migraine      | No             | No                                   | Low, 0.5               | p.G18R   | 0              | Waller et al., 2015               |
| 55      | Asian     | 1/9                       | IM                        | Frequent paroxysmal episodes of unsteadiness, headaches, nystagmus, vomiting. MRI normal. Present in unaffected father and brother | No             | Yes                                  | Normal                | p.T60M   | 0.00002         | Arsov et al., 2012               |
| 56      | British   | 8/28                      | IM                        | Myotonia and dystonia                                            | No             | No                                   | Normal                | p.G76V   | 0              | No                               |
| 57      | British   | 2/25                      | IF                        | PDE                                                                 | No             | No                                   | N/D                   | p.R91W   | 0              | Schneider et al., 2009            |
| 58      | British   | 6-13/18-78                | 2M 2F                     | PKD in three cases, PDE in one. Attacks typical of PKD           | Yes, family history of migraine. | No                                   | Normal                | p.R92W   | 0              | No                               |
| 59      | British   | 1 1/46                    | 3F                        | Severe PDE and PKD                                               | Yes, AD family history | No                                   | Low, 0.4               | p.M96V   | 0              | Leen et al., 2010                 |
| 60      | British   | Teens/49                  | IM 2F                     | PNDK                                                               | Affected mother | Yes                                  | Normal                | p.C201R  | 0              | No                               |
| 61      | British   | 8/24                      | IM                        | PND with epilepsy                                                 | No             | No                                   | N/A                   | p.R223W  | 0              | Leen et al., 2010                 |
| 62      | British   | 12/42                     | IM 1F                     | PDE                                                                | Dominant inheritance | Yes                                  | Normal                | p.A275T  | 0              | Weber et al., 2008                |
| 63      | British   | 15/28                     | IF                        | PDE and seizures                                                  | No             | No                                   | Low 0.55              | p.S285P  | 0              | No                               |
| 64      | Ireland   | 4/17                      | IM 2F                     | EA2, early absence seizures                                       | No             | No                                   | N/A                   | p.T295M  | 0              | Weber et al., 2008                |
| 65      | British   | Child/36                  | IM 2F                     | PDE                                                                | No             | No                                   | N/A                   | T295M    | 0              | Weber et al., 2008                |
| 66      | British   | 5/13                      | IF                        | PKD, long and frequent episodes of dystonia and unusual tongue dystonia. | No             | No                                   | N/A                   | p.R333Q + PRRT2 (p.R216H) | 0        | Schneider et al., 2009           |
| 67      | British   | 4/54                      | IM 1F                     | PDE, migraines and seizures                                       | No             | No                                   | Low, 0.5              | p.R333Q  | 0              | Schneider et al., 2009            |
| 68      | British   | 12/26                     | IM 1F                     | PDE, seizures                                                     | Daughter affected | Yes                                  | Low, 0.5              | p.R333W  | 0              | Wang et al., 2000                 |

AD = Alzheimer’s disease; EA = episodic ataxia; HM = hemiplegic migraine; N/D = not determined.
There was a spectrum of clinical features and many patients had additional clinical features such as seizures. The frequency of migraine and hemiplegic migraine was highly associated with these phenotypes although this is also common in the general population. Some individuals in the extended PKD families did not have a movement disorder at all or were affected by seizures or hemiplegic migraine alone. The usual mechanism for PRRT2 mutations is loss of function due to nonsense-mediated decay, leading to haploinsufficiency (Figs 2–5) and likely lead to a lack of SNAP25/SNARE interaction and increased vesicle release. Segregating PRRT2 missense mutations were also identified where there was no change in the PRRT2 mRNA, but we expect a loss of SNAP25/SNARE interaction or prevention of the PRRT2 protein from anchoring to the presynaptic membrane, and thus leading to a similar lack of inhibition of vesicle release due to reduced tethering (Fig. 5).

Fewer mutations were identified in the SLC2A1 and PNKD genes, and primarily in patients with PKD and PNKD (Fig. 1 and Table 2). The patients with SLC2A1 mutations had the broadest spectrum of clinical phenotypes. There was overlap clinically with PKD (as in the p.R223W family) and PNKD (as with the p.C210R family). This group were the most extensively investigated before a genetic diagnosis was sought, and fasting CSF glucose was frequently low in affected individuals with a more complex phenotype associated with seizures but usually normal with a movement disorder alone. There was also a greater rate of an incorrect clinical diagnosis and overlap with other channelopathies, as with the family with the p.G76V mutation and abnormal McManis testing, and in the family with the p.R333Q mutation and unusual tongue dystonia as part of the phenotype. These families are similar to those first described in 1892 as atypical Thomsen’s disease (Kure, 1892). The p.R333Q had an additional variant in the PRRT2 gene (p.P216H), which may be benign or modifying the effect of the p.R333Q mutation. In addition there was evidence of reduced penetrance in SLC2A1, most clearly in the family with the p.T60M mutation that presented with paroxysmal attacks, headaches and nystagmus where the father and brother had the mutation but were unaffected (see family tree, Fig. 2). The p.T60M mutation has previously been identified in idiopathic epilepsy, further extending the heterogeneity.

In the episodic ataxia cohort, one family was identified with a mutation in the PRRT2 gene, one with a defect in the SLC2A1 gene and two familial hemiplegic migraine families were identified, one with a PRRT2 mutation and one with a novel PNKD mutation. The familial hemiplegic migraine families were of most interest as they have a typical phenotype and the mutations segregate in the family. The novel PNKD mutation is a frameshift deletion located in exon 10, which is predicted to cause a truncated protein, this segregated with the disease, predicted pathogenic and was not identified in controls (Figs 2, 3 and 4B). Alternate splicing of the PNKD gene results in three isoforms of the protein of varying length; PNKD-S, PNKD-M (both expressed ubiquitously), and PNKD-L (expressed in the CNS) (Shen et al., 2011). All previously reported mutations are located in the 5’ end of the gene, found in both PNKD-L and PNKD-S but not PNKD-M. This mutation, instead affects PNKD-L and PNKD-M and the location and truncating effect of the change in shortening the PNKD protein is likely to lead to reduced RIM/RIM1 binding (Shen et al., 2015) in the SNARE complex and abnormal vesicle release (Fig. 5).

While there is a great deal more to be understood, it seems likely that these three paroxysmal genes are acting on the presynaptic terminal, possibly with overlapping pathways, and thus result in a similar dysregulated and possibly increased vesicular release. Although there is clinical overlap, there are also additional clinical features. This overlap is seen in the brain expression patterns where genes with a similar mechanism have identical regional expression patterns (Supplementary Fig. 1) as for PRRT2, SNAP25, KCNA1 and CACNA1A (all presynaptic) where they share highest expression levels in the cerebellum, and frontal, temporal and occipital cortices as compared with SLC2A1 and PNKD. This could explain the subtle phenotypic differences and the regional effect on vesicle release. It has recently been reported that overexpression of wild-type PNKD in rat hippocampal cultures reduced neurotransmitter release in comparison to an empty vector, whereas

**Table 3 Clinical phenotypes of the four PNKD probands and mutations**

| n   | Ethnicity | Age at onset/current age | Sex | Phenotypic description | Family history | Family members tested for segregation | Genetics | Frequency in ExAC | Previously reported phenotype | Previously reported (reference) |
|-----|-----------|--------------------------|-----|------------------------|----------------|----------------------------------------|----------|-------------------|--------------------------------|--------------------------------|
| 69  | German    | Teens/20s                | 3F 3M | PKD                    | Four generation large family | Yes | p.A7V | 0 | Lee et al., 2004; Rainier et al., 2004 | Rainier et al., 2004 |
| 70  | British   | 16/32                    | 2M 2F | PNKD with atypical features | Yes, father, paternal uncle and grandmother | Yes | p.A9V | 0 | Lee et al., 2004; Rainier et al., 2004 | Rainier et al., 2004 |
| 71  | British   | 8-22/20-64               | 17M 10F | PNKD                  | Several affected over three generations | Yes | p.A9V | 0 | Lee et al., 2004; Rainier et al., 2004 | Rainier et al., 2004 |
| 72  | British   | 30-34/44-78              | 2M 1F | Familial hemiplegic migraine | Father, great-uncle and proband over three generations | Yes | c.1022delC, p.Pro341fs | 0 | No | |
overexpression of mutant PNKD did not. This suggested that PNKD also has a role in regulating presynaptic exocytosis (Lee et al., 2015). It is also known that PRRT2 interacts with SNAP25, a protein important in facilitating synaptic exocytosis (Lee et al., 2015). Therefore, we suggest a possible disease mechanism whereby both PNKD and PRRT2 perform similar roles in restricting synaptic exocytosis. Disease-causing mutations that either reduce levels of PRRT2 or disrupt PNKD function reduce this restriction and result in excessive neurotransmitter release (Fig. 5). It is unclear how SLC2A1 mutations contribute to this theory, but it has been shown that they result in reduced glucose transport into the brain, so perhaps glucose is also involved in the regulation of exocytosis. The functional consequence of the regional expression patterns remains to be seen but may indicate that SLC2A1 and PNKD pathways are more closely related to dystonic genes located in the basal ganglia and brainstem.

Little is known about how disruption of these proteins results in migraine, a clinical manifestation that has been seen frequently here and elsewhere. However, in a recent study, transgenic mice with human monogenic migraine gene mutations (thus mimicking the types of migraine seen in this cohort) were shown to display increased glutamatergic neurotransmission and cerebral hyperexcitability (Ferrari et al., 2015). This finding indicates that the lack of neurotransmitter release regulation postulated here could also result in the migraine exhibited. There is clearly a large pathophysiological overlap between all of these related neurological disorders, which required further investigation to be understood more fully.

Overall this work reveals a wide spectrum of mutations and phenotypes and has expanded the broad phenotypic spectrum of these paroxysmal movement disorders, suggesting where possible, as part of the investigative work-up, all three genes should be analysed in these conditions. We also highlight novel mutations and a likely distinct mechanism for 3′ PNKD mutations that lead to PNKD-L dysregulation. There is genetic and phenotypic overlap amongst other episodic movement disorders with episodic ataxia, the neuronal channelpathies and familial hemiplegic migraine all being identified with defects in these three genes.

Acknowledgements

The authors would like to thank the patients and their families for their help.

Funding

This study was supported by the Medical Research Council (MRC UK) for the MRC Centre for Neuromuscular Diseases and project grant MR/J004758/1, Muscular Dystrophy UK, The Wellcome Trust in equipment and strategic award (Synaptopathies) funding (WT093205MA and WT104033AIA), The Brain Research Trust (BRT), The MSA Trust, the European Union Seventh Framework Programme (NeurOmics) and the National Institute for Health Research (NIHR) University College London Hospitals (UCLH) Biomedical Research Centre.

Supplementary material

Supplementary material is available at Brain online.

References

Agostinelli S, Traverso M, Accorsi P, Beccaria F, Belcastro V, Capovilla G, et al. Early-onset absence epilepsy: SLC2A1 gene analysis and treatment evolution. Eur J Neurol. 2013 May;20: 856–9.
Andrisse S, Koehler RM, Chen JE, Patel GD, Vallurupalli VR, Ratliff BA, et al. Role of GLUT1 in regulation of reactive oxygen species. Redox Biol 2014; 2: 764–71.
Arsov T, Mullen SA, Rogers S, Phillips AM, Lawrence KM, Damiano JA, et al. Glucose transporter 1 deficiency in the idiopathic generalized epilepsies. Ann Neurol 2012; 72: 807–15.
Bawazir WM, Gevers EF, Flatt JF, Ang AL, Jacobs B, Oren C, et al. An infant with pseudo-olygokalemia, hemolysis, and seizures: cation-leaky GLUT1-deficiency syndrome due to a SLC2A1 mutation. J Clin Endocrinol Metab 2012; 97: E987–93.
Benz R, Viecelli A, Taverna C, Schelosky L. Paroxysmal non-kinesigenic dyskinesia due to spinal cord infiltration of low-grade B cell non-Hodgkin’s lymphoma. Ann Hematol 2012; 91: 463–5.
Bhatia KP. Familial (idiopathic) paroxysmal dyskinesias: an update. Semin Neurol 2001; 21: 69–74.
Bhatia KP. Paroxysmal dyskinesias. Mov Disor 2011; 26: 1157–65.  
Bhatia KP. The paroxysmal dyskinesias. J Neurol 1999; 246: 149–55.  
Demirkiran M, Jankovic J. Paroxysmal dyskinesias: clinical features and classification. Ann Neurol 1995 Oct;38: 571–9.

References to funding

Cao L, Huang XJ, Zheng L, Xiao Q, Wang XJ, Chen SD. Identification of a novel PRRT2 mutation in patients with paroxysmal kinesigenic dyskinesias and c.649dupC as a mutation hot-spot. Parkinsonism Relat Disord 2012; 18: 704–6.
Chen WJ, Lin Y, Xiong ZQ, Wei W, Ni W, Tan GH, et al. Exome sequencing identifies truncating mutations in PRRT2 that cause paroxysmal kinesigenic dyskinesia. Nat Genet 2011; 43: 1252–5.
Cioffi R, Brunet N, Rudolf G, Massacrier A, Salmi M, Bataillard M, et al. PRRT2 links infantile convulsions and paroxysmal dyskinesia with migraine. Neurology 2012; 79: 2097–103.
de Vries B, Callenbach PM, Kamphorst JT, Weller CM, Koelwijn SC, ten Houten R, et al. PRRT2 mutation causes benign familial infantile convulsions. Neurology 2012; 79: 2154–5.
Engelen M, Tijssen MA. Paroxysmal non-kinesigenic dyskinesia in antiphospholipid syndrome. Mov Disor 2005; 20: 111–13.
Erro R, Sheerin UM, Bhatia KP. Paroxysmal dyskinesias revisited: a review of 500 genetically proven cases and a new classification. Mov Disor 2014; 29: 1108–16.
Ferrari MD, Klever RR, Terwindt GM, Ayata C, van den Maagdenberg AMJM. Migraine pathophysiology: lessons from mouse models and human genetics. Lancet Neurol 2015; 14: 65–80.

Friedman J, Olivera J, Silhavy JL, Gabriel SB, Glessen JG. Mild paroxysmal kinesigenic dyskinesia caused by PRRT2 missense mutation with reduced penetrance. Neurology 2012; 79: 946–8.

Friedman A, Zakrzewsk-Pniewska B, Domitz I, Lee HY, Pracek L, Kwicinska H. Paroxysmal non-kinesigenic dyskinesia caused by the mutation of MR-1 in a large Polish kindred. Eur Neurol 2009; 61: 39–41.

Fung EL, Ho YY, Hui J, Wong JH, Ng TB, Fong NY, et al. First report of GLUT1 deficiency syndrome in Chinese patients with novel and hot spot mutations in SLC2A1 gene. Brain Dev 2011; 33: 170–3.

Gardiner AR, Bhatia KP, Stamelou M, Dale RC, Kurian MA, Schneider SA, et al. PRRT2 gene mutations: from paroxysmal dyskinesia to episodic ataxia and hemiplegic migraine. Neurology 2012; 79: 2115–21.

Ghezzi D, Visconi C, Ferlini A, Gulandli F, Mereghetti P, DeGrandis D, et al. Paroxysmal non-kinesigenic dyskinesia is caused by mutations of the MR-1 mitochondrial targeting sequence. Hum Mol Genet 2009; 18: 1058–64.

Goksen S, Yilmaz S, Klepper J, Serdaroglu G, Tekgul H. Video/EEG recording of myoclonic absences in GLUT1 deficiency syndrome with a hot-spot R126C mutation in the SLC2A1 gene. Epilepsy Behav 2011; 21: 200–2.

Gowers W. Epilepsy and other chronic convulsive diseases: their causes. Symptoms Treat 1901; 1: 75–6.

Guerrini R, Mink JW. Paroxysmal disorders associated with PRRT2 mutations shake up expectations on ion channel genes. Neurology 2012; 79: 2086–8.

Hashimoto N, Kagitani-Shimono K, Sakai N, Otomo T, Tominaga K, Guerrini R, Mink JW. Paroxysmal disorders associated with PRRT2 mutations cause benign familial infantile epilepsy and infantile convulsions with choreoathetosis syndrome. Am J Hum Genet 2012; 90: 152–60.

Ishii A, Yasumoto S, Ibara Y, Inoue T, Fujita T, Nakamura N, et al. Genetic analysis of PRRT2 mutations cause benign familial infantile epilepsy and infantile convulsions with choreoathetosis syndrome. Am J Hum Genet 2012; 90: 152–60.

Kedera P, Xiao J, Puschmann A, Momcilovic D, Wu SW, LeDoux MS. Novel PRRT2 mutation in an African-American family with paroxysmal kinesigenic dyskinesia. BMC Neuro 2012; 12: 93.

Heron SE, Grinton BE, Kivity S, Afasti Z, Zuberi SM, Hughes JN, et al. PRRT2 mutations cause benign familial infantile epilepsy and infantile convulsions with choreoathetosis syndrome. Am J Hum Genet 2012; 90: 152–60.

Kortes A. Paroxysmal kinesigenic choreoathetosis: an entity within the paroxysmal choreoathetosis syndrome. Description of 10 cases, including 1 autopsy. Neurology 1967; 17: 680–90.

Kinali M, Junghuth H, Unson LH, Sewry CA, Manzur AY, Mercuri E, et al. Expanding the phenotype of potassium channelopathy: severe neuromyotonia and skeletal deformities without prominent Episodic Ataxia. Neuromuscul Disord 2004; 14: 689–93.

Kure S. Atypical Thomsen's disease. Tokyo Igakukai Zasshi. J Tokyo Med Assoc 1892; 6: 505–14.

Kurian MA, Wang Y, Kaluger R, Minks J, Chelly J, et al. Quality control parameters on a large dataset of regionally expressed genes. J Hum Genet 2011; 56: 846–51.

Lee HY, Xu Y, Ahn AH, Auhberger GW, Pandolfo M, et al. The gene for paroxysmal non-kinesigenic dyskinesia encodes an enzyme in a stress response pathway. Hum Mol Genet 2004; 13: 3161–70.

Lee HY, Fu YH, Pracek LJ. Episodic and electrical nervous system disorders caused by nonchannel genes. Annu Rev Physiol 2015; 77: 525–41.

Leen WG, Klepper J, Verbeek MM, Leferink M, Hofste T, van Engelen BG, et al. Glucose transporter-I deficiency syndrome: the expanding clinical and genetic spectrum of a treatable disorder. Brain 2010; 133: 655–70.

Li H-F, Chen W-J, Ni W, Wang K-Y, Liu G-L, Wang N, et al. PRRT2 mutation correlated with phenotype of paroxysmal kinesigenic dyskinesia and drug response. Neurology 2013; 80: 1534–5.

Li J, Zhu X, Wang X, Sun W, Feng B, Du T, et al. Targeted genomic sequencing identifies PRRT2 mutations as a cause of paroxysmal kinesigenic choreoathetosis. J Med Genet 2012; 49: 76–8.

Liu Q, Qi Z, Wan XH, Li JY, Shi J, Lu Q, et al. Mutations in PRRT2 result in paroxysmal dyskinesias with marked variability in clinical expression. J Med Genet 2012; 49; 79–82.

Lombroso CT, Fischman A. Paroxysmal non-kinesigenic dyskinesia: pathophysiological investigations. Epileptic Disord 1999;1: 187–93.

Mao C-Y, Shi C-H, Song B, Wu J, Ji Y, Qin J, et al. Genotype-phenotype correlation in a cohort of paroxysmal kinesigenic dyskinesia cases. J Neurol Sci 2014; 340: 91–3.

Mount L, Reback S. Familial paroxysmal choreoathetosis. Arch Neurol 1940; 44: 841–7.

Muhle H, Helbig I, Froslev TG, Suls A, von Spiczak S, Klatten LL, et al. The role of SLC2A1 in early onset and childhood absence epilepsy. Epilepsia Res 2013; 105: 229–33.

Ono S, Yoshuura K, Kinoshita A, Kikuchi T, Nakane Y, Kato N, et al. Mutations in PRRT2 responsible for paroxysmal kinesigenic dyskinesias also cause benign familial infantile convulsions. J Hum Genet 2012; 57: 338–41.

Pons R, Cuenca-Leon E, Miravit E, Pons M, Xaidara A, Yourouskos S, et al. Paroxysmal non-kinesigenic dyskinesia due to a PNKD recurrent mutation: report of two Southern European families. Eur J Paediatr Neurol 2012; 16: 86–9.

Rainier S, Thomas D, Tokarz D, Ming L, Bui M, Plein E, et al. Myofibrillogenesis regulator 1 gene mutations cause paroxysmal dystonic choreoathetosis. Arch Neuro 2004; 61: 1025–9.

Raskind WH, Bolin T, Wolff J, Fink J, Matsushita M, Litt M, et al. Further localization of a gene for paroxysmal dystonic choreoathetosis to a 5-cM region on chromosome 2q34. Hum Genet 1998; 102: 93–7.

Schneider SA, Paisan-Ruiz C, Garcia-Gorostiaga I, Quinn NP, Weber YG, Lerche H, et al. GLUT1 gene mutations cause sporadic paroxysmal exercise-induced dyskinesias. Mov Disor 2009; 24: 1684–8.

Scheffer IE, Grinton BE, Heron SE, Kivity S, Afasti Z, Iona X, et al. PRRT2 phenotype spectrum includes sporadic and febrile-related infantile seizures. Neurology 2012; 79: 2104–8.

Shen Y, Ge WP, Li Y, Hirano A, Lee HY, Rohlmann A, et al. Protein mutated in paroxysmal dyskinesia interacts with the active zone protein RIM and suppresses synaptic vesicle exocytosis. Proc Natl Acad Sci USA 2015; 112: 2935–41.

Shen Y, Lee HY, Rawson J, Ojha S, Babbutt P, Fu YH, et al. Mutations in PNKD causing paroxysmal dyskinesia alters protein cleavage and stability. Hum Mol Genet 2011; 20: 2322–32.

Silveira-Moriyama L, Gardiner AR, Meyer E, King MD, Smith M, Rakshi K, et al. Clinical features of childhood-onset paroxysmal kinesigenic dyskinesia with PRRT2 gene mutations. Dev Med Child Neurol 2013; 55: 327–34.

Sohn YH, Lee PH. Paroxysmal choreoathetoid disorders. In: Vinken PJ, Bruyn GW, editors. Handbook of clinical neurology. Vol. 100. Philadelphia, PA: Elsevier; 2011. pp. 367–73.

Specchio N, Terracciano A, Trivisano M, Cappelletti S, Claps D, Travaglini L, et al. PRRT2 is mutated in familial and non-familial benign infantile convulsions. Eur J Hum Genet 2009; 17: 77–81.

Suls A, Dedeken P, Godijn K, Van Esch H, Dupont P, Cassiman D, et al. PRRT2 mutations cause benign familial infantile convulsions and episepse is due to mutations in SLC2A1, encoding the glucose transporter GLUT1. Brain 2008; 131(Pt 7): 1831–44.

Trabzuni D, Ryten M, Walker R, Smith C, Imran S, Ramasamy A, et al. Quality control parameters on a large dataset of regionally
dissected human control brains for whole genome expression studies. J Neurochem 2011; 119: 275–82.

van Rootselaar AF, Schade van Westrum S, Velis DN, Tijssen MA. The paroxysmal dyskinesias. Pract Neurol 2009; 9: 102–9.

Vercueil L. Paroxysmal non-kinesigenic dyskinesia. Epileptic Disord 2000; 2: V–VI.

Vermeer S, Koolen DA, Visser G, Brackel HJ, van der Burgt I, de Leeuw N, et al. A novel microdeletion in 1p34.2p34.3, involving the SLC2A1 (GLUT1) gene, and severe delayed development. Dev Med Child Neurol 2007; 49: 380–4.

Wang D, Kranz-Eble P, De Vivo DC. Mutational analysis of GLUT1 (SLC2A1) in Glut-1 deficiency syndrome. Hum Mutat 2000; 16: 224–31.

Wang JL, Cao L, Li XH, Hu ZM, Li JD, Zhang JG, et al. Identification of PRRT2 as the causative gene of paroxysmal kinesigenic dyskinesias. Brain 2011; 134(Pt. 12): 3493–501.

Weber MB. Familial paroxysmal dystonia. J Nerv Ment Dis 1967; 145: 221–6.

Weber YG, Storch A, Wuttke TV, Brockmann K, Kemptle J, Maljevic S, et al. GLUT1 mutations are a cause of paroxysmal exertion-induced dyskinesias and induce hemolytic anemia by a cation leak. J Clin Invest. 2008; 118: 2157–68.

Weller CM, Leen WG, Neville BG, Duncan JS, Vries BD, Geilenkirchen MA, et al. A novel SLC2A1 mutation linking hemiplegic migraine with alternating hemiplegia of childhood. Cephalalgia 2015; 35: 10–15.

Wu L, Tang H-D, Huang X-J, Zheng L, Liu X-L, Wang T, et al. PRRT2 truncated mutations lead to nonsense-mediated mRNA decay in Paroxysmal Kinesigenic Dyskinesia. Parkinsonism Relat Disord. 2014; 20: 1399–404.

Zorzi G, Conti C, Erba A, Granata T, Angelini L, Nardocci N. Paroxysmal dyskinesias in childhood. Pediatr Neurol 2003; 28: 168–72.