Identification of 3 Novel Patients with *CLCN2*-Related Leukoencephalopathy due to *CLCN2* Mutations

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Dear Sir,

CLC-2 is a chloride channel, which plays an important role in brain ion and water homeostasis both in mice and humans [1]. *CLCN2*-related leukoencephalopathy (CC2L) is a recently identified rare disorder, caused by autosomal recessive mutations in *CLCN2*, which results in CLC-2 chloride channel dysfunction and myelin microvacuolization with characteristic brain MRI features of leukoencephalopathy [2]. We report three patients with CC2L, who carry genetically verified *CLCN2* mutations and were presented between 1999 and 2014 to our clinic.

Patients

All patients were women with adult-disease onset (Table 1). Patient 1 presented with gait-speech difficulty and had dysarthria, nystagmus, ataxic gait and dysmetria. Patient 2 presented with headache, imbalance and blurry vision. She had diplopia, parosmia and ataxic gait. Patient 3 presented with right-side numbness and ataxic gait.

All laboratory tests were normal. Patient 2 had an abnormal visual evoked po-

tential, which showed unilateral P100 latency increase, although the rest of the ophthalmologic examination was normal. All patients had cerebellar ataxia in common and showed distinct MRI findings of bilateral T2-hyperintensities and diffusion restriction in the posterior limbs of the internal capsules, midbrain cerebral peduncles and middle cerebellar peduncles, and low signal in the same localizations on apparent diffusion coefficient (ADC) maps (Fig. 1). After verifying *CLCN2* (NM_004366.5) mutations (Table 1), they were diagnosed with CC2L and had stable or very slowly progressive disease courses.

Discussion

The prevalence of CC2L is not known. Sixteen patients are identified till date, since its discovery in 2013 [2, 3], but there could be more undiagnosed patients existing, who possibly remain asymptomatic. The frequency of the disease should be interpreted cautiously due to the autosomal recessive nature of CC2L, the relatively high consanguinity in our country and our

clinic being a referral center. As an ideal approach, a multicenter MRI database may identify potential individuals with CC2L, who could further be evaluated for genetic mutations.

CC2L is characterized by mild and non-specific neurological involvement, visual impairment and male infertility [4, 5]. Mild cerebellar ataxia with action tremor-gait instability is the most common neurological finding, and patients are reported to remain ambulatory during follow-up [2]. The highly variable clinical presentations and age of onset may be partially related to the extent of CLC-2 loss of function [2]. However, the current knowledge on CC2L from the small number of affected individuals does not favor any genotype-phenotype correlations [3]. In line with this, patients 2 and 3 may be expected to have a larger impact on the molecular function of their CLC-2 channels due to the mutations identified, but they had milder clinical phenotypes compared with patient 1. The correlation between the functional impact of the identified mutations and clinical phenotypes needs to be investigated further as more patients with CC2L are identified.

Fig. 1. Brain MRI of patient 1: signal abnormalities in the posterior limbs of the internal capsules (**a–c**), midbrain cerebral peduncles (**d–f**) and middle cerebellar peduncles (**g–i**). Hyperintensities

on axial T2-weighted images (**a, d, g**), diffusion restriction on axial diffusion-weighted images (**b, e, h**) and corresponding low signal on axial apparent diffusion coefficient maps (**c, f, i**).

Major inclusion criteria for brain MRI are abnormally high signal on T2-weighted images and abnormally low signal on T1-weighted images in the posterior limbs of the internal capsules, midbrain cerebral peduncles and middle cerebellar peduncles [3]. MRI shows diffusion restriction and

low ADC values [2] in the same structures as a result of myelin microvacuolization [3]. In adult patients, the diffusion restriction is observed in the specific white matter structures, whereas in pediatric patients there is relatively a more diffuse diffusion restriction [2].

The differential diagnosis for CC2L depends mostly on bilateral abnormal MRI findings, since clinical features are usually nonspecific. The list primarily includes autosomal-dominant adult onset leukodystrophy related to *LMNB1* duplications, CMT1X – gap junction beta-1 (*GJB1*) gene

Table 1. Demographics and clinical features

	Patient 1	Patient 2	Patient 3
Age/sex	47/ Woman	33/ Woman	47/ Woman
Consanguineous parents	+	+	–
Early psychomotor development and cognitive level	Normal	Normal	Normal
Disease onset age (year)	28	27	46
Initial clinic presentation	1999	2013	2014
Presenting signs	Gait difficulty, imbalance	Imbalance, headache, blurry vision	Numbness, imbalance
Vision acuity	Normal	Normal	Normal with eyeglasses
Visual fields	Normal	Normal	Normal
Nystagmus	+	–	–
Hearing loss	Unilateral mild conductive type hearing loss	Normal	Normal
Ataxia	Moderate	Mild	Mild
<i>CLCN2</i> (NM_004366.5) mutations	Homozygous c.1412G>A, p.Arg471His mutation MM	Homozygous p.Glu475LysfsTer79 FSM	Compound heterozygous p.Leu435ArgfsTer7 FSM

MM= missense mutation; FSM= frame shift mutation.

mutation encoding connexin 32 [6], and Pelizaeus-Merzbacher like disease (PMLD) [3].

In conclusion, the MRI findings are essential for the diagnosis of *CLCN2*-related leukoencephalopathy, a novel and rare disorder caused by *CLCN2* mutations with nonspecific clinical findings, distinct MRI patterns and further features yet to be discovered.

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Statement of Ethics

Informed consent form was obtained from each individual.

Disclosure Statement

The authors have no conflicts of interest to declare.

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