Epileptic phenotypes related to the UNC79-UNC80-NALCN protein complex

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Abstract

The NALCN gene encodes a voltage-independent, non-selective, cation channel permeable to Na+, K+ and Ca2+ which forms a large channel complex with two other proteins, products of UNC80 and UNC79 genes.

The UNC80 gene is essential for the stability and function of the NALCN sodium leak channel and for bridging NALCN to UNC79 to form a functional complex.

This channel complex is mainly expressed in the central nervous system (CNS) and it plays a crucial role in regulating the resting membrane potentials and neuronal excitability.

Mutations in the NALCN gene have recently been linked to human diseases: two autosomal-recessive conditions (IHPRF1, Infantile hypotonia with psychomotor retardation and characteristic facies; INAD; Infantile neuroaxonal dystrophy with facial dysmorphisms and skeletal anomalies) and one autosomal-dominant condition (CLIFHADD; Congenital contractures of the limbs and face, hypotonia and developmental delay). Homozygous or compound heterozygous mutation in the UNC80 gene are associated to the IHPRF2 syndrome (Infantile hypotonia with psychomotor retardation and characteristic facies-2). In addition to facial dysmorphism and psychomotor retardation, epilepsy is often associated with IHPRF1 and IHPRF2 Syndrome but the epileptic phenotype has never been accurately described.

“Recently two novel mutations in NALCN gene have been observed in two Sardinian siblings with IHPRF1 Syndrome and epilepsy. Here we describe the epileptic phenotype of these two Sardinian siblings and compare their findings with those previously reported in NALCN and UNC80 recessive mutated patients.”

KEY WORDS: UNC79-UNC80-NALCN protein complex, NALCN mutations, UNC80 mutations, IHPRF, epilepsy.

Introduction

The NALCN, UNC80 and UNC79 genes encode proteins that form a channel complex (1). UNC80 bridges NALCN to UNC79, and these 2 genes are involved in the folding, stabilization, cellular localization and activation of NALCN essential for channel function (2, 3).

The NALCN gene encodes a voltage-independent, nonselective, cation channel permeable to Na+, K+ and Ca2+ and belongs to the 24-transmembrane domain ion channel superfamily. These channels have four homologous domains consisting of six transmembrane helices separated by three cytoplasmatic linkers. The homologous domains participate in the formation of the pores.

NALCN gene is expressed mainly in the CNS (dentate gyrus and pyramidal cell layer of the hippocampus, granular and Purkinje cell layers and dentate nucleus of cerebellum), but is also been found at a lower level in the heart, lymph nodes, pancreas, thyroid (4). This
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The UNC79-UNC80-NALCN protein complex plays a crucial role in regulating resting membrane potentials and neuronal excitability. Mutations in the NALCN gene have recently been linked to human diseases: IHPRF1 (Infantile hypotonia with psychomotor retardation and characteristic facies), an autosomal-recessive condition (3-7); CLIFHADD (Congenital contractures of the limbs and face, hypotonia and developmental delay), an autosomal-dominant condition (5, 8); and INAD (Infantile neuroaxonal dystrophy) with facial dysmorphism and skeletal anomalies (9), an autosomal-recessive condition. IHPRF2 Syndrome has been associated to recessive mutations of UNC80 gene (2, 10-12).

UNC79 so far has no corresponding human phenotype (2), although mouse knockout models for NALCN and UNC79 both fail to nurse and die shortly after birth.

So far only 4 families with 11 cases have been reported in literature with NALCN recessive mutations (6, 7, 9) and 10 families with a total of 19 cases have been found with UNC80 recessive mutations (2, 10-12). Epilepsy was reported both in patients with NALCN and UNC80 mutations.

Seizures, usually generalized, tended to be well controlled by AED (antiepileptic drugs).

Recently two novel bi-allelic truncating mutations in NALCN have been detected by Angius et al in two Sardinian siblings and they represent the fifth family reported worldwide with IHPRF1 syndrome (13).

We describe the epileptic phenotype of these two Sardinian siblings and compare them with NALCN and UNC80 recessive mutated patients.

Case report

The two siblings are a 22 years old male and a 20-years-old female born from non consanguineous parents.

Clinical and epileptic phenotype of the two siblings are summarized in Table 1.

Male patient was born at the 34\(^{\text{th}}\) pregnancy week with a birth weight of 1790 g, while his sister was born at term with adequate birth weight.

They both show dysmorphic features: triangular face, bitemporal narrowing, high nasal bridge, downslanting palpebral fissures, posterior rotated low set-ears, large and persistently opened mouth, long fingers. Since the first months of life hypotonia and developmental delay were evident. Now they can maintain sitting position without support but they are unable to walk, never developed speech, present a severe intellectual disability and a severe sleep disturbance. Their ophthalamic picture is characterized by convergent strabismus, lack of fixation, micropapilla, smaller retinal vessels and peripheral corioretinal sclerosis.

Both of them present severe hyperkinetic movement disorder and traits of autism spectrum disorder are evident in the male patient. Because of feeding difficulties, during his first months of life, the male patient underwent PEG system implantation, lately removed because of swallowing improvement. The female patient was fed through nasogastric tube until age of 8 months. As for her brother, later she had an improvement, but they both showed poor growth and actually have a very low body weight. Severe chronic constipation is also present.

In the male patient, epilepsy onset was at the age of 4 years. Seizures were generalized and polymorphic: first atypical absences and later myoclonic-atonic, tonic and tonic-clonic seizures.

EEG shows disorganized background activity, high amplitude multifocal and diffuse slow/sharp waves and spike-wave discharges intermingled with fast anterior activity (Fig. 1).

At present, despite politherapy (valproic acid, ethosuximide, rufinamide and clonazepam) and EEG improvement, he experiences weekly tonic and tonic-clonic seizures, mainly during sleep.

The female patient experienced the first seizure at 6 years. Seizures were generalized and polymorphic: first atypical absences and later myoclonic-atonic. At onset, EEG showed disorganized background activity both in awake and sleep state, high amplitude diffuse and multifocal slow/sharp waves, irregular spikes and spike-wave discharges (Fig. 2).

The last EEGs are characterized by irregular low voltage theta activity intermingled with diffuse high voltage theta activity.

Since the introduction of valproic acid and ethosuximide, she is seizure free.

Their metabolic and genetic workups were normal. MRI showed diffuse atrophy and cerebellar hypoplasia.


Discussion

IHPRF syndrome is due to recessive mutations in the NALCN gene, but some cases with similar phenotype are associated with UNC80 mutations. Until now, the syndrome associated with NALCN mutation was described only in four families worldwide (6, 7). Neurological picture is quite similar in NALCN and UNC80 recessive mutated patients, while dysmorphic faces, autism and hyperkinetic movement disorders seem to be more frequent in UNC80 patients.

The two Sardinian siblings show clinical features more similar to the latter patients. Among patients with recessive mutations, epilepsy is reported in 5/9 (6, 7). In the first Saudi Arabia family, seizures are not semiologically described but it is reported a low frequency and good response to pharmacological therapy; in the second family from Israel, seizures were generalized tonic-clonic in both sisters.
with onset at about four years and good response to vigabatrin, but one of them later developed resistant status epilepticus and died for aspiration pneumonia and respiratory failure. Epilepsy is also reported in 2 patients with INAD and dysmorphic face associated with \textit{NALCN} mutation (9) and in 9 patients with \textit{UNC80} mutation (10-12).

In both groups, seizures are usually generalized (atypical absences, myoclonic-atonic, tonic and tonic-clonic) and only one patient experienced neonatal...
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Figure 1 - Sleep EEG patient 1.

Figure 2 - Sleep EEG patient 2.
focal seizures. Response to therapy is usually good. EEG characteristics of patients with IHPRF Syndrome have never been accurately described. An intermittent activity with slow background, without epileptic activity was reported for some NALCN mutated patients (6). Most UNC80 patients show disorganized background activity, diffuse slow/sharp waves usually without epileptic activity, but four of them present epileptic activity (spike-wave) and rarely multifocal anomalies (10-12). The two Sardinian siblings experienced generalized polymorphic seizures and their age onset of epilepsy is similar to that reported for NALCN patients (6, 7), while the UNC80 patients usually have a earlier onset during the first months of life (11, 12). Their EEG is disorganized and bi-frontal and diffuse high amplitude sharp/slow-waves can be seen. Particularly, EEG of the male patient has a high frequency of epileptic discharges and, despite a recent moderate improvement, his electric activity is much more impaired than his sister. AED response has been good in the female patient with seizure freedom, while the male patient still has weekly seizures. Epileptologic characteristics of these two Sardinian patients seem to be much more similar to that of UNC80 mutated patients because of seizure semiology and EEG pattern. Based on the potential implication of the UNC79-UNC80-NALCN channel complex in human physiology and diseases, NALCN could represent an attractive candidate gene to be tested in individuals with infantile encephalopathy with epilepsy but further investigations are needed to clarify its role.

References