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"Real world effectiveness of cerliponase alfa in classical and atypical patients. A case series"

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ABSTRACT

Introduction: Late infantile neuronal ceroid lipofuscinosis is an autosomal recessive disease caused by mutations in the CLN2/*TPP1* gene, with secondary enzyme deficiency. In classical phenotypes, initial symptoms include seizures and delayed language development between 2 and 4 years of age. This article describes the presentation of CLN2 disease in a cohort of Colombian patients, as well as the impact of treatment on the course and progression of the disease.

Methods: Case series report of 8 patients with a confirmed diagnosis of neuronal ceroid lipofuscinosis treated with cerliponase alfa who remained on clinical and paraclinical follow-up for up to 24 months before and after treatment.

Results: An atypical phenotype, associated with initial symptoms and late diagnosis, was present in 5/8 patients. The most frequent symptoms were seizures and developmental delay, with age of onset at 24 months (classical phenotype) and 48 months (atypical phenotype). A novel mutation (c.1438G > A) was found in two siblings. All of the patients received cerliponase alfa, and there were no serious adverse events. No decline in the clinical status greater than 2 points on Hamburg, Weill Cornell and CNL2 clinical assessment scale was observed during follow-up after treatment initiation.

Conclusion: This is the first case series reported for neuronal ceroid lipofuscinosis patients in Colombia. In contrast with other reports, the majority of cases reported here displayed an atypical phenotype. Our study highlights the importance of early diagnosis and timely initiation of therapy, which is a feasible therapy, well tolerated by patients and accepted by caregivers in this country, generating a positive impact in the quality of life of CLN2 patients and on disease outcome.

1. Introduction

Neuronal ceroid lipofuscinoses (NCLs) are lysosomal storage diseases that, collectively, represent the most common group of infantile encephalopathies [1]. Late infantile neuronal ceroid lipofuscinosis type 2 (CLN2) (OMIM#204500) [2], is an autosomal recessive disease caused by mutations in the CLN2/*TPP1* gene located on chromosome 11p15.4 which encodes the tripeptidyl-peptidase 1 (TPP1) enzyme [3]. To date, a total of 131 independent mutations of the CLN2/*TPP1* gene have been reported, with a phenotypical description in 65%, and 66% of these mutations being familiar genotypes [4]. The most common mutations are c.509-1G > C and c.622C > T [p.Arg208X] which, taken together, account for 60% of the mutational spectrum of the disease, with genotype depending on the geographic region [4].

Worldwide, the reported incidence is 0.1–7/100.000 and varies by geographic region [5,6]. In Latin America in 2015, after a 12-year follow-up, the integrated program in Argentina reported a total of 22 patients from different Latin American countries [7]. To date, there are no reports on the incidence of this disease in Colombia.

The classical phenotype of the disease manifests in a previously healthy child. The initial symptoms vary: tonic-clonic or partial seizures (70% of cases) and delayed language development (57% of cases) with

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age of onset between 2 and 4 years [8,9]. Progressive motor and cognitive decline occur later, leading to the loss of voluntary movement, altered swallowing and speech (6 years), visual loss due to retinal atrophy (between 7 and 10 years) and death in mid adolescence (10–15 years) [8]. There are several variants of the classical phenotype, classified as atypical, which are characterized by late onset with variable clinical manifestations [10,11].

Diagnosis is confirmed by the presence of compatible homozygote or compound heterozygote mutations of the CLN2/*TPP1* gene and TPP1 enzyme deficiency or absence [11]. Electroencephalographic (EEG) evidence of early photoparoxysmal response may be observed, starting at low frequencies (1–2 Hz), with irregular activity, attenuated background activity and epileptiform abnormalities in the occipital region [11]. Brain nuclear magnetic resonance imaging (MRI) may show early evidence of progressive cerebral and cerebellar atrophy which is a key diagnostic finding suggestive of CLN2. Other findings include: reduced grey matter volume and hyperintense periventricular white matter [12]. As part of the ophthalmological assessment, optic coherence tomography may show evidence of retinal degeneration [13] and accumulation of hyperreflective material [14], abnormal visual evoked potentials and diminished electroretinographic amplitudes [14].

There are multiple disease-specific scales that measure progression of the disease and allow assessment of the degree of disability based on different characteristics such as motor and visual function, gait and language (Table 1).

Patients with CLN2 disease require multidisciplinary management in order to address symptoms, such as distress, maintain motor function and improve/maintain quality of life depending on the clinical status at the time of diagnosis.

Recombinant human TPP1 (cerliponase alfa) is the only available disease-modifying therapy and has been shown to slow the progression of decline in the motor-language domains of the CLN2 Rating Scale, when compared to patients receiving symptomatic management alone in the study of the natural history of the disease [18]. This article describes the presentation of CLN2 in a cohort of Colombian patients, as well as the impact of the treatment on the course and progression of the disease.

2. Materials and methods

We present a case series report of 8 patients from three regions of Colombia (Bogotá, Villavicencio and Armenia); patients were seen in four centers (Fundación Hospital Pediátrico la Misericordia, Hospital Central de la Policía Nacional, Hospital Universitario Clínica San Rafael in Bogotá; and Neuroconexión in Armenia) and followed by a multidisciplinary team for a time period of 24 months. Patients had a diagnosis of CLN2, of classical or atypical phenotypical expression, confirmed by biochemical and enzymatic testing and genotyping of the CLN2/*TPP1* gene. Atypical patients were those with enzymatic and/or molecular diagnosis of CLN2 who exhibited a protracted or milder clinical course, with variation in age at onset, rate of progression and clinical manifestation, as compared to the classical late-infantile phenotype [19]. Enzyme replacement therapy was offered in accordance to local regulatory guidelines for patients with confirmed diagnosis of CLN2, aged 3–15 years with motor-language Hamburg score within 3–6 [20].

Data was collected retrospective based on clinical charts with a prospective follow-up after treatment. Patient variables included in accordance with the protocols for clinical record reviews at each of the centers: a) Descriptive variables (age, gender, affected relatives, consanguinity and perinatal history); b) Variables pertaining to the onset of the disease (age of onset, initial symptoms, symptoms on first visit, initial pharmacological management, initial paraclinical testing and *TPP1* sequencing); c) Follow-up variables (additional symptoms over the course of the disease, scores on the CLN2 Clinical Rating Scale, Weill Cornell scale, Hamburg scale and Hamburg motor and language scale); d) Variables pertaining to enzyme replacement therapy (starting age, dose, frequency, time on therapy, adherence percentage), and variables related to the course of the disease after treatment (scores on the CLN2 Clinical Rating Scale, Weill Cornell scale, Hamburg motor and language scales).

An Excel (version 16.35) database was built including the variables described above, and statistical analysis was performed using the SPSS software package (version 26). Descriptive and inferential statistics were applied. Alfa error of 0.05, beta error of 80%, and a 95% confidence interval (CI) were established, with a statistically significant p value <0.05. Non-parametric tests (Wilcoxon) were performed given the non-normal sample distribution. Verification of 100% of the data was carried out for quality control in order to avoid information bias. Data anonymization was applied in order to ensure confidentiality, considering the sensitive nature of the information.

This article is in compliance with the Nuremberg Code of 1947 [21], the Universal Declaration of Human Rights of 1948 [22], Resolution 8430 of 1993 [23] and the Declaration of Helsinki [22]. Ethics Committee of Fundación Hospital Pediátrico la Misericordia (HOMI) (No. CEI-259-20) granted authorization for the study and signed informed consents for the use of the information was obtained from the guardians of each of the patients.

3. Results

This case series report considered 8 patients with a phenotypical diagnosis of CLN2 with enzymatic, biochemical and *TPP1* genotyping (Annex 1). An atypical phenotype was found in 5/8 of the patients. There were two pairs of siblings in the cohort of patients, both families having the atypical phenotype: One of the patients received diagnostic confirmation while still asymptomatic because of the history of an older sibling, who was previously identified.

At the time of the first visit, 7/8 of the patients presented with seizures, 6/8 with language abnormalities, 4/8 with developmental regression, and 3/8 with ataxia and movement disorders. The initial symptoms by subgroups are shown in Table 2, the most predominant being seizures in both the classical (3/3 and 5/5) and the atypical phenotypes (4/5). During clinical follow-up up to the time of diagnosis, and before enzyme replacement therapy, disease progression continued as illustrated in the table.

Medians and respective inter-quartile ranges for age of symptom onset, age at first visit, age of developmental regression, time from diagnosis to initiation of therapy, age at treatment initiation, and time on therapy are described in Table 3; 6/8 of the patients had an affected relative and none of the patients had a history of consanguinity.

During follow-up after the first visit and before the start of enzyme replacement therapy, 3/8 patients developed ataxia, 3/8 developed movement disorders and 2/8 regressed in their language development. In the subgroup analysis, among patients with the classical phenotype 2/3 patients (developed ataxia, 2/3 patients developed movement

Table 1
Tools for clinical follow-up of patients with CLN2.

Tool	Measurement	Author and validation
Hamburg Scale	Motor, language and visual assessment Total score 9 (3 for each category). Separate item for grand mal seizures, not taken into account in the total score	Steinfeld, [15]
Weill Cornell Scale	Swallowing, gait, motor function and language assessment Total score 12 (3 for each category)	Worgall, [16]
CNL2 clinical assessment scale	Motor, language assessment, and combination of the two The combined total score is 6.	Wyrwich, [17]

Table 2
Qualitative clinical characteristics of the patients before initiating enzyme replacement therapy.

		Initial Symptoms		Symptoms prior enzyme replacement therapy	
		Classical ($n = 3$)	Atypical ($n = 5$)	Classical ($n = 3$)	Atypical ($n = 5$)
Symptoms	Asymptomatic	0	1	0	1
	Language abnormalities	3	3	3	3
	Movement disorders	1	1	3	2
	Ophthalmological alterations	0	0	3	1
	Ataxia	1	2	3	3
	Seizures	3	4	3	4
	Developmental regression	1	3	3	3
Additional tests	EEG (abnormal)	3	3		
	Electroretinogram (abnormal)	3	0		
	Visual evoked potentials (abnormal)	0	1		
	Photoparoxysmal response (abnormal)	0	1		
	Brain MRI (abnormal)	3	2		
	Optic tomography (abnormal)	2	0		

Table 3
Quantitative clinical characteristics.

Case series			Classical		Atypical	
	Median	Range (Q1-Q3)	Median	Range (Q1-Q3)	Median	Range (Q1-Q3)
Age of onset of symptoms (months)	36	(30–48)	24	[24–30]	48	(36–48)
Age at the time of the first visit (months)	48	(30–66)	30	[30–33]	60	(60–84)
Age of developmental regression (years)	3	(2–10)	3	(3–3)	4.5	(2–10)
Time from diagnosis to therapy initiation (months)	11	(3–13)	4	(3–11)	13	(5–13)
Age at treatment initiation (months)	72	(48–192)	52	(48–62)	139	(72–192)
Time on therapy (months)	9	(7–24)	16	(15–24)	8	(7–9)
Time from onset of symptoms until diagnosis (months)	12	(11–12)	6	(5–6)	24	(21–27)

disorders, 2/3 patients developed developmental regression, and 3/3 patients developed ophthalmological alterations. On the other hand, among patients with atypical phenotypes, 1/5 patient developed ataxia, 1/5 patient developed movement disorder, and 1/5 patient developed ophthalmological alterations, while one patient remained asymptomatic; the other findings were unchanged (Table 2). The EEG, MRI and other additional tests of patients with the classical and the atypical phenotyes are described in Table 2. Levels of tripeptyl-peptidase 1 enzyme activity were undetectable or below the normal range in 8/8 of the patients in both groups. Table 4 shows the data for CLN2/*TPP1* gene sequencing.

As part of the treatment received before starting on enzyme replacement therapy, 6/8 of the patients were receiving valproic acid, 2/8 levetiracetam, 3/8 clobazam, 1/8 oxcarbazepine and 1/8 lamotrigine (Table 5).

Table 5
Treatment prior and post enzyme replacement therapy.

	Initial treatment		Additional treatment post enzyme replacement therapy	
	Classical ($n = 3$)	Atypical ($n = 5$)	Classical ($n = 3$)	Atypical ($n = 5$)
Valproic acid	3	3	3	1
Clobazam	3	0	3	1
Lamotrigine	0	1	0	0
Levetiracetam	1	1	1	2
Oxcarbazepine	1	0	1	0

Table 4
TPP1 sequencing.

Phenotype	Allele	Location	Nucleotide change	Aminoacid change	Clinical significance	Contig position (GRCh38p7)	Reference
Atypical	Compound heterozygous	Exon 12 Intron 8	c.1438G > A / c.1076-2A > T	p.Val480Met/ NA	NA / Probably pathogenic	6614979C > T / 6,616,076 T > C	NA / Caillaud et al., 1999
Atypical	Compound heterozygous	Exon 12 Intron8	c.1438G > A / c.1076-2A > G	p.Val480Met/ NA	NA / Probably pathogenic	6614979C > T / 6,616,076 T > C	NA / Caillaud et al., 1999
Atypical	Compound heterozygous	Exon 6 Intron 7	c.622C > T / c.887-10A > G	p.Art208*/ Varia	Pathogenic / Conflicting	6,617,040 G > A/ 6,616,513 T > C	Sleat et al., 1999/ Noher de Halac et al., 2005
Atypical	Compound heterozygous	Exon 6 Intron 7	c.622C > T / c.887-10A > G	p.Art208*/ Varia	Pathogenic / conflicting	6,617,040 G > A/ 6,616,513 T > C	Sleat et al., 1999/ Noher de Halac et al., 2005
Atypical	Compound heterozygous	Intron 8 Intron 7	c.1076-2A > T/ c.887-10A > G	NA/ Varia	Probably pathogenic/ Conflicting	6,616,076 T > C/ 6,616,513 T > C	Caillaud et al., 1999/ Noher de Halac et al., 2005
Classical	Compound heterozygous	Intron 8 Exon 6	c.1076 -2A > T/ c.616C > T	NA/p. arg206Cys	Probably pathogenic/ Pathogenic	6,616,076 T > C/ 6,617,046 G > A	Caillaud et al., 1999/ Kousi et al., 2012
Classical	Compound heterozygous	Exon 5 Intron 8	c.471C > A / c.1076-2A > G	p.Tyr157*/NA	Probably pathogenic / Probably pathogenic	6,617,338C > A /6616076 T > C	Sheth et al. 2018/ Caillaud et al., 1999
Classical	Compound heterozygous	Intron 8 Exon 6	c.1076-2A > T/ C·616C > T	NA/ p. arg206cys	Probably pathogenic/ Pathogenic	6,616,076 T > C/ 6,617,046 G > A	Caillaud et al., 1999/ Kousi et al., 2012

Table 6
Functional scales before and after treatment.

Functional scales		Case series	Classical phenotype	Atypical phenotype
		Median (Range)	Median (Range)	Median (Range)
Before treatment	CLN2 Clinical Rating Scale	4 (3–4)	2 (2–3)	4 (4–5)
	Weill-Cornell scale	8 (6–10)	6 (6–7)	8 (5–10)
	Hamburg scale	9 (7–10)	6 (5–7)	10 (9–11)
	Hamburg Motor and Language (ML) scale	4 (3–4)	3 (3–4)	4 (4–5)
After treatment	CLN2 Clinical Rating Scale	4 (2–4)	2 (2)	4 (4–5)
	Weill-Cornell scale	7 (5–10)	5 (5)	8 (5–10)
	Hamburg scale	9 (7–10)	10 (9–11)	7 (6–7)
	Hamburg ML scale	4 (3–4)	3 (3–4)	4 (4–5)

All of the patients received intracerebroventricular cerliponase alfa 300 mg every 15 days as enzyme replacement therapy, including one asymptomatic patient with enzymatic activity below the detection range. Median time from diagnosis to initiation of therapy was 11 months (IQR 3–13). Median time from treatment initiation until the study cut-off point (June 2020) was 9 months (IQR 7–24). Subgroup analysis revealed that, compared to patients with the classical phenotype, patients with atypical phenotypes showed older median age of onset (139 months vs. 52 months), longer median time from onset of symptoms until diagnosis (24 months vs. 6 months), longer median time from confirmation of diagnosis to initiation of treatment (13 months vs. 4 months), and shorter time on therapy (8 months vs. 16 months) (Table 3).

The analysis after initiation of enzyme replacement therapy with cerliponase alfa showed evidence of reduction in the use of valproic acid in the atypical phenotype patients (3/5 vs. 1/5), while there was increased use of levetiracetam (1/5 vs. 2/5) and clobazam (0/5 vs. 1/5). Medication use remained constant in the classical phenotype (Table 5).

Functional scores were analyzed before and after treatment initiation, as shown in Table 6. The statistical analysis did not show evidence of statistically significant differences between pre and post-treatment scores ($p > 0.005$).

The median time on enzyme replacement therapy was 9 months. Fig. 1 shows median scores on the CLN2 and Hamburg scales before and after treatment. Fig. 2 shows individual patient scores on the CLN2 scale during follow-up before and after treatment initiation.

Data from both atypical and classical phenotypes. Boxes show median values (middle lines) with a 25th and 75th percentiles; whiskers show values within two-thirds of the IQR. CLN2 = Late-infantile neuronal ceroid lipofuscinosis type 2.

Longitudinal follow up (in weeks) of CLN2 scale on individual patients according to their clinical phenotype.

During follow-up, there was no evidence of progression, defined as 2-point decline in the CLN2 scale (Fig. 3).

Data from both atypical and classical phenotypes. Bars show changes from baseline (prior to treatment) in CLN2 score during follow up. No progression was evident (defined as > -2 in CLN2 scale).

4. Discussion

To date, multiple genetic variants responsible for CLN2 disease have been described, with variable population allele frequencies. On CLN2/*TPP1* gene sequencing, the most frequent allele (6 of 16) in our case series was c.1076-2A > T (rs1424116749), an intronic variant in which the base replacement in region 3′ changes the CLN2/*TPP1* gene splicing acceptor [25]. It has been described as a potentially pathogenic variant and, moreover, it was present in all patients with the classical phenotype, accompanied by a second allele with a pathogenic mutation due to an amino acid (c.616C > T) or a stop codon (c.471C > A) replacement (Table 3). None of these mutations correspond to the most frequent genotypes in patients with CLN2 in the world [4], demonstrating the genetic heterogeneity and population variation of genetic diseases such as CLN2. This also reinforces the difference between the mutational profile in the Latin American population and profiles reported in other regions of the world.

In patients with the atypical phenotype, the most frequent allele variant (3 of 10) was c.887-10A > G (seen on the two siblings in the study), an intronic variant described for the first time in Portugal [26] and reported as the most frequent variant on the largest cohort described [27]. The authors postulate that the protein with this mutation could have residual activity. A nonsense mutation was also found in two patients (c.622C > T) while the c.1076-2A > G mutation described previously (altered splicing) was found in a third patient. The two remaining patients had one allele with a splicing alteration and another allele with a mutation not previously described in the literature (c.1438G > A), the clinical meaning of which has not been described. The assumption could be that, given its association with an atypical phenotype, it might have a partial functionality in the resulting

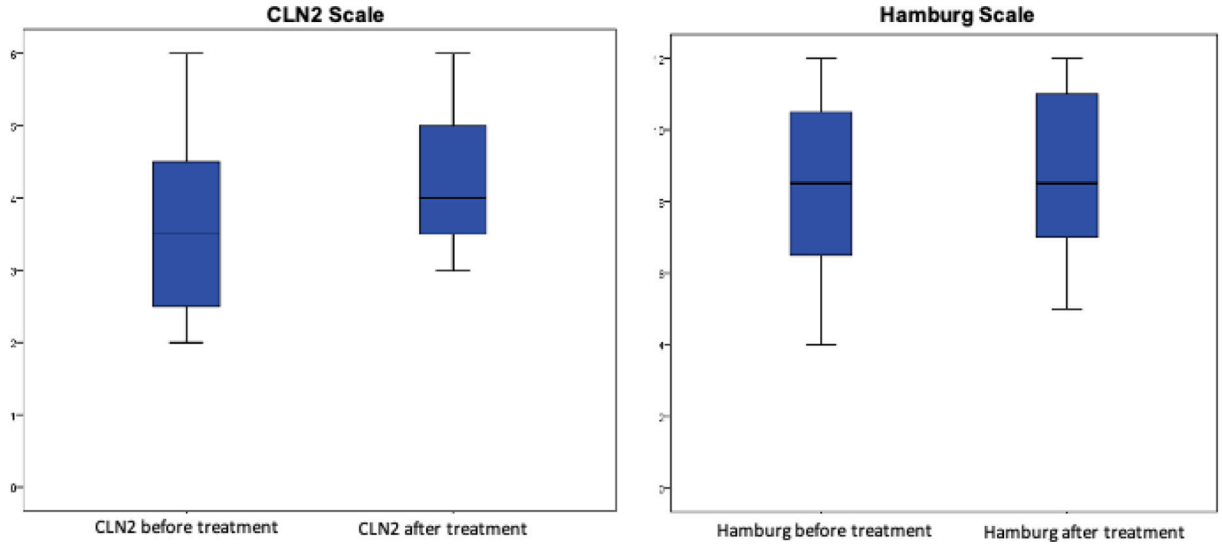

Fig. 1. Box and whisker plots before and after treatment on the CLN2 and Hamburg scales.

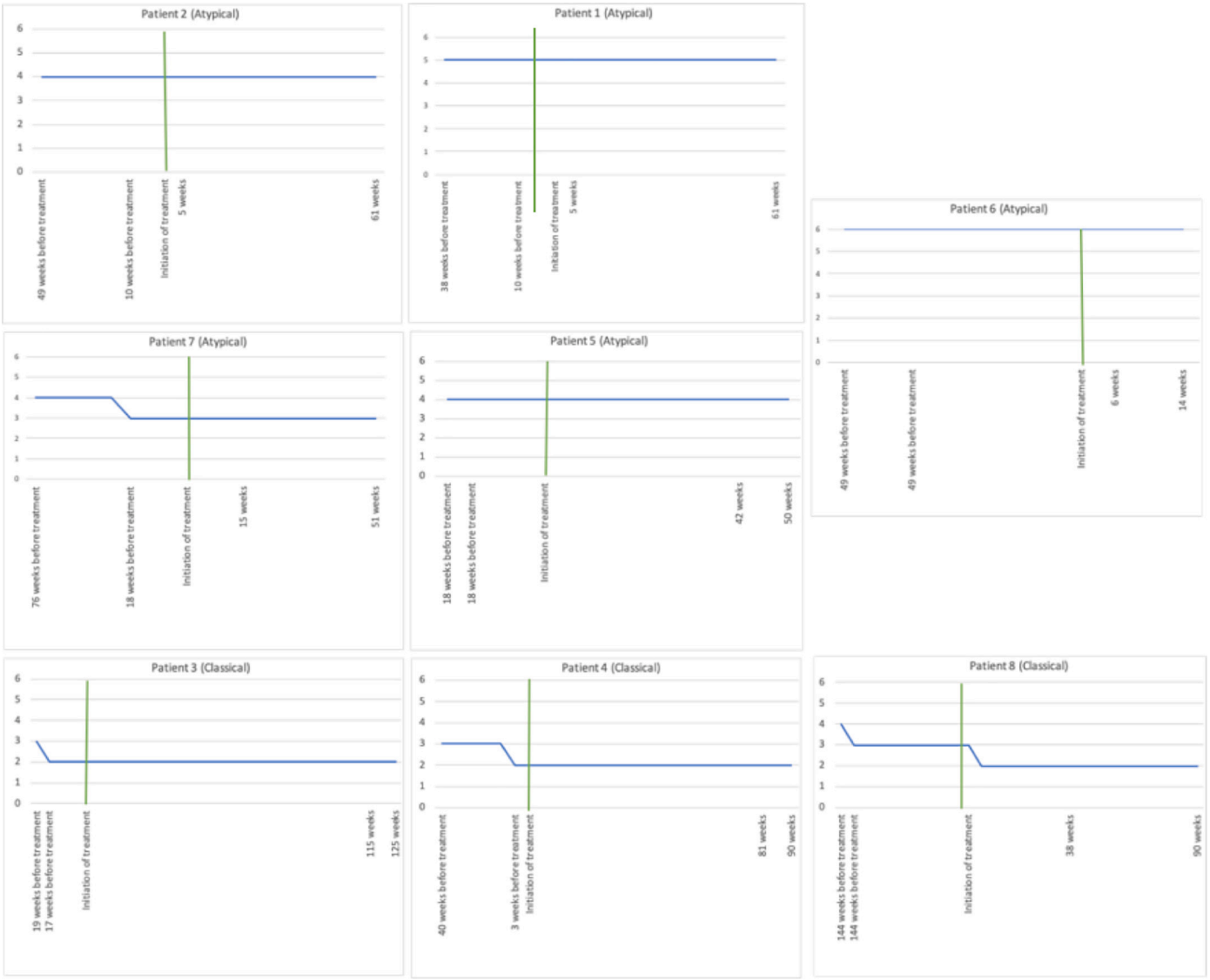

Fig. 2. Individual follow-up results for the case series according to clinical phenotype.

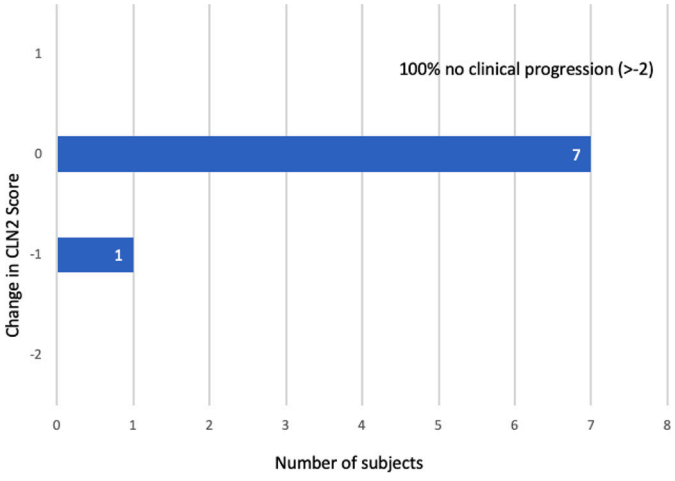

Fig. 3. Changes from baseline (prior to treatment) on the CLN2 scale during follow-up.

transcript. Genomic variations in atypical phenotypes support the hypothesis of a combination of intronic and nonsense/missense mutations associated with residual enzyme activity, allowing for a phenotype with a milder presentation, as previously reported [28].

In our patients, the classical phenotypical presentation is consistent with the evidence described in the literature [14,29,30], with a median age of onset of 24 months at which 3/3 showed seizure activity and regression in terms of language development. In the atypical phenotype, the median age of presentation was 48 months, with a broader age range (36–120 months) and seizures as the predominant initial symptom (4/5), followed by language alterations and developmental regression (3/5). A later course of the disease and a less severe phenotype is seen when the two subgroups are compared, which is consistent with the descriptions of atypical phenotypes [7,10]. The median interval from the onset of symptoms to the first visit was 3 years longer in the atypical phenotype patients, while time to diagnosis was 1.5 years longer, as well as time to initiation of enzyme replacement therapy (4 months in the classical phenotype vs. 13 months in the atypical phenotype). This may be explained by clinical variations and the later onset of symptoms in the atypical phenotype, which pose a diagnostic challenge, as evidenced in our study. Therefore, it is important to consider the possibility of CLN2 disease in patients with seizures and altered language development or

cognitive decline [7,10,11], particularly considering that the atypical phenotype was found in 5/8 of the patients, showing its high frequency in the Colombian population with CLN2. This is in contrast with the world reports of sporadic familial cases (7 patients in 5 families) [6,10] but is consistent with the findings in a Latin American cohort of 32 patients with atypical phenotype (unpublished data).

There are few publications to date reporting the real-world effectiveness of the cerliponase alfa in patients with CLN2 disease. During follow-up before initiating enzyme replacement therapy, marked decline was observed in the patients with the classical phenotype, 3/3 of whom developed ataxia, movement disorders, global developmental regression, ophthalmological alterations, with persistent seizures and language alterations. This is in contrast to a less severe course in the group of patients with the atypical phenotype, among whom 1/5 developed ataxia, 1/5 developed movement disorders, and 1/5 developed ophthalmological disorders. It is also important to mention the presence in this group of a patient who remained asymptomatic throughout the entire follow-up period, but who was diagnosed because of the affected sibling. At present, this patient is receiving treatment due to having undetectable TPP1 levels, although all the other clinical variables remained unchanged during follow-up.

All of the patients received enzyme replacement therapy with intracerebroventricular cerliponase alfa 300 mg every 15 days. Adherence to treatment varied from 58 to 100%: this variation can be attributed mainly to administrative hurdles or the decision of the caregivers, and was not secondary to adverse events. Greater administrative barriers that limited the administration of the medication were identified in the case of patient 8. Additionally, 50% of patients exhibited no seizures following the initiation of enzyme replacement therapy; this was unrelated to changes in the use of anticonvulsants. This is consistent with quality-of-life scales after the initiation of treatment with cerliponase alfa, with reported reductions in epileptic seizures [31]. Likewise, EEG progression was not present, whereas only one patient showed progression in MRI during follow-up (Annex 1).

The clinical status of the patients was compared before and after treatment using the Hamburg and Weill Cornell functional scales and the CLN2 Clinical Rating Scale. The comparative analysis of these scales (Figs. 1–4) showed no evidence of progression greater than −2 points in 100% of the sample; progression of −1 point was found in one patient (patient 8). These data support stabilization in the progression of the disease in both, the atypical and classical phenotypes, with an adequate safety profile that was similar to that reported in other studies [18,32,33], and specifically on atypical patients, where ERT is well tolerated and motor and language function stabilize in 79% of the patients and improvement in 7% [34].

5. Conclusions

Here we report the first case series of classical and atypical patients with CLN2 in Colombia. Evidence was found of a novel mutation in two patients with different allele frequency variants than those reported in the world literature. Evidence was also found of a higher number of patients with the atypical phenotype as compared to the classical phenotype in this case series, in contrast with international reports. However, the clinical characteristics identified in the two groups are consistent with those described previously in the literature. There is a prolonged latency between the onset of symptoms and diagnosis, particularly in the atypical phenotype, reflecting the detection challenge in patients of this region. In the CLN2 disease patients included in this case series, treatment with enzyme replacement therapy resulted so far, in no progression of motor and language decline, as determined using scales for clinical reassessment during follow-up. This study is among the first to report real-world evidence of effectiveness of the medication, with similar results to those reported in the world literature and a similar response to the use of cerliponase alfa in patients with both the classical and the atypical phenotypes. Additionally, our study highlights the importance of early diagnosis and timely initiation of therapy, which is a feasible therapy, well tolerated by patients and accepted by caregivers in this country, generating a positive impact in the quality of life of CLN2 patients and on disease outcome.

Ethics committee

This study was approved by the Ethics Committee of Fundación Hospital Pediátrico la Misericordia (HOMI), Bogotá, Colombia (No. CEI-259-20). A signed informed consent for publication of the clinical cases, laboratory results, paraclinical tests, photographs and videos was obtained from the children's parents.

Editorial coordination

Integralis HGS (Doctor Daniel Rodríguez and María Stella Salazar).

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Declaration of competing interest

Doctors Naranjo, Tavera, Mancilla and Espitia have received conference fees from BioMarin Pharmaceuticals Inc. Dr. Hernández has no disclosures to make.

Appendix A. Annex 1

Clinical phenotype	Atypical phenotype	Atypical phenotype	Atypical phenotype	Atypical phenotype	Atypical phenotype	Classical phenotype	Classical phenotype	Classical phenotype
ID	#1	#2	#5	#6	#7	#3	#4	#8
Natural	Bogota	Bogota	Bogota		Armenia	Bogota	Bogota	Bogota
Gender	Male	Female	Male	Female	Female	Female	Male	Male
Age	6	14	10	2	16	6	5	3
Age of onset	3	10	4		4	2	3	2
Age at first visit to pediatric neurology	5	12	7	2	5	2,5	3	2.5
Affected relatives	Yes	Yes	Yes	Yes	No	No	Yes	Yes
Initial symptoms	Seizure	Ataxia	Language regression	Asymptomatic Asymptomatic	Language regression	Language regression	Seizure	Global developmental delay

(continued on next page)

(*continued*)

Clinical phenotype	Atypical phenotype	Atypical phenotype	Atypical phenotype	Atypical phenotype	Atypical phenotype	Classical phenotype	Classical phenotype	Classical phenotype
ID	#1	#2	#5	#6	#7	#3	#4	#8
Symptoms at first visit	Seizure, Abnormal movements and language abnormalities	Seizure and developmental regression	Seizure, Ataxia and developmental regression		Seizure and developmental regression	Seizure and language abnormalities	Seizure and language abnormalities	Seizure, ataxia, developmental regression, abnormal movements and language
Course of the disease before treatment	No changes	Symptoms seen on initial visit accompanied by ataxia	Symptoms seen on initial visit accompanied and abnormal movements	Asymptomatic	Symptoms seen on initial visit accompanied by ataxia, abnormal movements and			ophthalmological alterations.
Symptoms seen on initial visit accompanied by ataxia, developmental regression and								ophthalmological alterations.
Symptoms seen on initial visit accompanied by ataxia, r developmental egression, abnormal movements and								ophthalmological alterations.
Symptoms seen on initial visit accompanied by ataxia, abnormal movements and								ophthalmological alterations.
Additional tests before treatment	EEG: No abnormalities Brain MRI: No abnormalities	EEG: Abnormal Brain MRI: Abnormal	EEG: Abnormal Brain MRI: Abnormal					Photoparoxysmal response: Abnormal Visual potentials: No abnormalities Electroretinogram: No abnormalities
EEG: No abnormalities Brain MRI: No abnormalities Visual potentials: No abnormalities	EEG: Abnormal Brain MRI: No abnormalities Visual potentials: Abnormal							
EEG: Abnormal Brain MRI: No abnormalities. Visual potentials: No abnormalities								Electroretinogram: Abnormal Optic coherence tomography: Abnormal
EEG: Abnormal Brain MRI: Abnormal Visual potentials: No abnormalities								Electroretinogram: Abnormal Optic coherence tomography: Abnormal
EEG: Abnormal Brain MRI: Abnormal Visual potentials: No abnormalities								Electroretinogram: Abnormal
Tripeptidyl-peptidase 1 nmol/h/ml	Undetectable	Undetectable	<1.8	<1.6	0.7	Undetectable	0.3	0.6
Additional tests during treatment	EEG with no changes		Brain MRI with no changes. EEG with no changes	Brain MRI with no changes.	Brain MRI Abnormal with evidence of progression. EEG with no changes	Brain MRI with no changes. EEG with no changes	Brain MRI with no changes. EEG with no changes	
Initial treatment		Valproic acid	Valproic acid and Levetiracetam		Valproic acid and Lamotrigine	Valproic acid Levetiracetan and Clobazam	Valproic acid Clobazam	Valproic acid Clobazam and Oxcarbazepine
	6	13	10		16	4	4	5

(*continued on next page*)

(*continued*)

Clinical phenotype	Atypical phenotype	Atypical phenotype	Atypical phenotype	Atypical phenotype	Atypical phenotype	Classical phenotype	Classical phenotype	Classical phenotype
ID	#1	#2	#5	#6	#7	#3	#4	#8
Age of enzyme replacement therapy (Cerliponase alfa) initiation								
Months on enzyme replacement therapy (Cerliponase alfa)	9	9	7		7	24	15	16
Adherence to enzyme replacement therapy (Cerliponase alfa)	100%	100%	92%		62%	58%	80%	74%
Treatment associated to Cerliponase alfa		Valproic acid	Levetiracetam		Clobazam Levetiracetam	Clobazam Levetiracetam Valproic acid	Clobazam Valproic acid	Clobazam Oxicarbazepine Valproic acid
Last epileptic seizure during post-treatment follow-up	36	25	13		0	17	12	10
Enzyme replacement therapy (Cerliponase alfa)-related adverse event		Hyperpyrexia	Pleocytosis	Pleocytosis		Seizure		

References

[1] A. Jalanko, Y.T. Braulke, Neuronal ceroid lipofuscinoses, Biochim Biophys Acta - Mol Cell Res [internet] 1793 (4) (2009) 697–709. Available at: https://doi.org/10.1016/j.bbamcr.2008.11.004.

[2] Online Mendelian Inheritance in Man O. Johns Hopkins University, Baltimore, MD. [Internet]. Número OMIM: 204500, Available at: https://www.omim.org/entry/204500, 2020.

[3] D.E. Sleat, R.J. Donnelly, H. Lackland, C.G. Liu, I. Sohar, R.K. Pullarkat, et al., Association of mutations in a lysosomal protein with classical late-infantile neuronal ceroid lipofuscinosis, Science 277 (5333) (1997) 1802–1805.

[4] E. Gardner, M. Bailey, A. Schulz, M. Aristorena, N. Miller, Y.S.E. Mole, Mutation update: review of TPP1 gene variants associated with neuronal ceroid lipofuscinosis CLN2 disease, Hum Mutat [internet] 40 (11) (2019) 1924–1938. Available at: https://doi.org/10.1002/humu.23860.

[5] S.J. Moore, D.J. Buckley, A. MacMillan, et al., The clinical and genetic epidemiology of neuronal ceroid lipofuscinosis in Newfoundland, Clin. Genet. 74 (3) (2008) 213–222.

[6] R.E. Williams, H.R. Adams, M. Blohm, et al., Management strategies for CLN2 disease, Pediatr. Neurol. 69 (2017) 102–112.

[7] R. Kohan, F. Pesaola, N. Guelbert, P. Pons, A.M. Oller-Ramírez, G. Rautenberg, et al., The neuronal ceroid lipofuscinoses program: a translational research experience in Argentina, Biochim Biophys Acta - Mol Basis Dis [internet] 1852 (10) (2015) 2301–2311. Available at: https://doi.org/10.1016/j.bbadis.2015.05.003.

[8] R.E. Williams, L. Aberg, T. Autti, H.H. Goebel, A. Kohlschütter, Y.T. Lönnqvist, Diagnosis of the neuronal ceroid lipofuscinoses: an update, Biochim Biophys Acta - Mol Basis Dis. 1762 (10) (2006) 865–872.

[9] M. Nickel, A. Simonati, D. Jacoby, et al., Disease characteristics and progression in patients with late-infantile neuronal ceroid lipofuscinosis type 2 (CLN2) disease: an observational cohort study, Lancet Child Adolesc Heal. 2 (8) (2018) 582–590.

[10] M. Elleder, L. Dvořáková, L. Stolnaja, H. Vlášková, H. Hålková, R. Druga, et al., Atypical CLN2 with later onset and prolonged course: a neuropathologic study showing different sensitivity of neuronal subpopulations to TPP1 deficiency, Acta Neuropathol. 116 (1) (2008) 119–124.

[11] M. Fietz, M. AlSayed, D. Burke, J. Cohen-Pfeffer, J.D. Cooper, L. Dvořáková, et al., Diagnosis of neuronal ceroid lipofuscinosis type 2 (CLN2 disease): expert recommendations for early detection and laboratory diagnosis, Mol. Genet. Metab. 119 (1–2) (2016) 160–167.

[12] M.J. Bennett, The neuronal ceroid-lipofuscinoses, Dev Disabil Res Rev. 17 (3) (2013) 254–259.

[13] A. Orlin, D. Sondhi, M.T. Witmer, M.M. Wessel, J.G. Mezey, S.M. Kaminsky, et al., Spectrum of Ocular Manifestations in CLN2-Associated Batten (Jansky-Bielschowsky) Disease Correlate with Advancing Age and Deteriorating Neurological Function, PLoS One 8 (8) (2013) e73128, https://doi.org/10.1371/journal.pone.0073128 (eCollection 2013).

[14] M. Chang, J.D. Cooper, B.L. Davidson, et al., The neuronal ceroid lipofuscinosis (Batten Disease). En: Mole, S. E. y R. E. G. H. Williams, editor. CLN2, Oxford University Press, Oxford, 2011, pp. 80–109.

[15] R. Steinfeld, P. Heim, H. von Gregory, K. Meyer, K. Ullrich, H.H. Goebel, et al., Late infantile neuronal ceroid lipofuscinosis: quantitative description of the clinical course in patients with CLN2 mutations, Am. J. Med. Genet. 112 (4) (2002) 347–354.

[16] S. Worgall, M.V. Kekatpure, L. Heier, D. Ballon, J.P. Dyke, D. Shungu, et al., Neurological deterioration in late infantile neuronal ceroid lipofuscinosis, Neurology. 69 (6) (2007) 521–535.

[17] K.W. Wyrwich, A. Schulz, M. Nickel, P. Slasor, T. Ajayi, D.R. Jacoby, et al., An adapted clinical measurement tool for the key symptoms of CLN2 disease, J Inborn Errors Metab Screen. 6 (2018), 232640981878838.

[18] A. Schulz, T. Ajayi, N. Specchio, E. de los Reyes, P. Gissen, D. Ballon, et al., Study of intraventricular cerliponase alfa for CLN2 disease, N. Engl. J. Med. 378 (20) (2018) 1898–1907.

[19] N. Specchio, N. Pietrafusa, M. Trivisano, Changing times for CLN2 disease: the era of enzyme replacement therapy, Ther. Clin. Risk Manag. 16 (2020) 213–222, https://doi.org/10.2147/TCRM.S241048.

[20] Ministerio de Salud y de Protección Social, Instituto Nacional de Vigilancia de Medicamentos y Alimentos (INVIMA). Resolución No. 201942117 de 24 de Septiembre de, 2019.

[21] J.A. Mainetti, Código de Núremberg, in: Ética Médica 1, 1947, p. 2.

[22] Organización de las Naciones Unidas para la Educación la Ciencia y la Cultura Unesco, Declaración Universal de los Derechos Humanos. Mineduc.cl [Internet], 1948, p. 6. Available at: http://www.mineduc.cl/usuarios/ccalificacion/doc/201205101657450.declaracion_universal_de_los_derechos_humanos.pdf.

[23] Ministerio de Salud. Resolución 8430 de, Ministerio de Salud y Protección Social, República de Colombia 1993 (1993) 1–19.

[24] S. Holm, Declaration of Helsinki, in: En: International Encyclopedia of Ethics [Internet], 2013, https://doi.org/10.1002/9781444367072.wbiee230. Available at:.

[25] National Center for Biotechnology Information, NM_000391.4(TPP1):c.1076-2A>T [Internet]. ClinVar [consultado el 10 de marzo del 2019]. Available at: https://www.ncbi.nlm.nih.gov/clinvar/variation/555109/, 2018.

[26] C. Bessa, C.A. Teixeira, A. Dias, M. Alves, S. Rocha, L. Lacerda, et al., CLN2/TPP1 deficiency: the novel mutation IVS7-10A>G causes intron retention and is associated with a mild disease phenotype, Mol. Genet. Metab. 93 (1) (2008) 66–73.

[27] C.M. Lourenço, A. Pessoa, C.C. Mendes, C. Rivera-Nieto, D. Vergara, M. Troncoso, et al., Revealing the clinical phenotype of atypical neuronal ceroid lipofuscinosis type 2 disease: Insights from the largest cohort in the world, J Paediatr Child Health (2020) 1–7, https://doi.org/10.1111/jpc.15250 (October).
[28] R. Kohan, M.N. Carabelos, W. Xin, K. Sims, N. Guelbert, et al., Neuronal ceroid lipofuscinosis type CLN2: a new rationale for the construction of phenotypic subgroups based on a survey of 25 cases in South America, Gene. 516 (1) (2013) 114–121.
[29] M. Nickel, A. Schulz, A. Kolhlschütter, et al., Late language acquisition and unexplaied epilepsy are indicators of easily detectable CLN2 disease, Eur J Paediatr Neurol. 19 (S38) (2015).
[30] S. Worgall, M.V. Kekatpure, L. Heier, et al., Neurological deterioration in late infantile neuronal ceroid lipofuscinosis, Neurology. 69 (2007) 521–535.
[31] Biomarin Pharmaceutical (Canada) Inc, Clinical Review Report CERLIPONASE ALFA (BRINEURA) [Internet]. Cadth Common Drug Review, Available at: https://www.cadth.ca/sites/default/files/cdr/clinical/SR0430_KalydecoR117H_CL_Report.pdf, 2019.
[32] A. Schulz, N. Specchio, P. Gissen, E. de los Reyes, H. Cahan, P. Slasor, et al., Persistent Treatment Effect of Cerliponase Alfa in Children with CLN2 Disease: A 3 Year Update from an Ongoing Multicenter Extension Study, Abstr 45th Annu Meet Soc Neuropediatr 50 (2019) 2020.
[33] E. Wibbeler, R. Wang, E. de los Reyes, N. Specchio, P. Gissen, N. Guelbert, et al., Cerliponase Alfa for the Treatment of Atypical Phenotypes of CLN2 Disease: A Retrospective Case Series, J Child Neurol. (2020), https://doi.org/10.1177/0883073820977997.
[34] B. Estublier, A. Cano, C. Hoebeke, S. Pichard, D. Scavarda, I. Desguerre, et al., Cerliponase alfa changes the natural history of children with neuronal ceroid lipofuscinosis type 2: the first French cohort, Eur J Paediatr Neurol [Internet]. 30 (2021) 17–21, https://doi.org/10.1016/j.ejpn.2020.12.002.