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Clinical and Electrophysiological Aspects in Children with Spinal Muscular Atrophy Type 1, 2 and 3 before Treatment

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Abstract

Aim: Analysis of electrophysiological changes in children with spinal muscular atrophy (SMA) type 1, 2 and 3 before starting specific treatment and establishing clinical-electrophysiological correlations.

Patients and method: Retrospective study of patients with SMA type 1, 2 and 3 genetically confirmed between 1-16 years of age. The relationship between the amplitude of the Compound Muscle Action Potential (CMAP) and the type of disease, respectively the number of SMN2 copies, as well as the correlations with the scores on the functional scales applied according to the SMA form (The Children's Hospital of Philadelphia Infant Test of Neuromuscular Disorders/CHOP INTEND and Hammersmith Functional Motor Scale Expanded/HFMSE) were followed.

Results: A significant correlation was demonstrated between CMAP and HFMSE for type 2 SMA ($p < 0.01$, $r = 0.687$), a good statistical correlation between CMAP and CHOP for type 1 SMA ($p < 0.05$, $r = 0.586$). Needle electromyography (EMG) showed active denervation in both early (1st, 2nd) and late (3rd) forms, demonstrating the ongoing degenerative process, along with elements of collateral reinnervation much more evident in the late form (3rd).

Conclusions: Retrospective analysis of the group of patients without specific treatment demonstrated electrophysiological changes suggestive of peripheral motoneuron degenerative damage. The decrease in CMAP amplitude is associated with the SMA type and the scores on the functional scales.

Keywords: Spinal Muscular Atrophy; Electromyography; Functional Assessment

Introduction

Spinal muscular atrophy (SMA) is a degenerative neuromuscular disorder with autosomal recessive genetic transmission characterized by progressive loss of spinal and brainstem motor neurons, causing amyotrophy, motor deficit, swallowing and respiratory disorders [1,2]. Most cases of SMA are caused by the biallelic mutation in the SMN1 gene (telomeric) at the cr 5q level (homozygous deletion) [3]. There are also rare situations with the compound heterozygous variant or punctiform mutations. The SMN2 gene (centromeric) also present on chromosome 5 contributes to a small extent to the production of SMN protein and thus to the phenotypic aspect of the disease [4].

The aim of this study was the analysis of a representative group of patients with SMA before initiating treatment, with the assessment of clinical and electrophysiological parameters and subsequently with their monitoring in the evolution of the disease. It is a retrospective study, for the first time in Romania, applied to children with type 1, 2 and 3 of SMA.

The proposed objectives are the to establish statistical correlations between CMAP amplitude and scores on functional motor scales (CHOP and HFMSE). We will also follow the relationship between the clinical appearance and the number of SMN2 copies, between the electromyographic appearance (needle EMG) and the type of disease, between the CMAP value in patients with SMA/healthy children of the same age.

Based on previous studies that considered electrophysiological parameters (CMAP) as markers for disease progression and prognosis [5,6], we formulated the working hypothesis that within the proposed group of patients of CNCRNC there will be a correlation with statistical significance between CMAP and the scores on the motor scales corresponding to the type of disease.

Classification

Four types of spinal muscular atrophy are described in the pediatric population: type 0 (prenatal) with onset during intrauterine life, decreased fetal movements, severe generalized hypotonia, severe respiratory failure, joint contractions, facial diplegia, atrial septal defect; type 1 (infantile) characteristic of the infant, with the onset of symptoms from birth to 3 months, severe form with severe hypotonia, breathing and swallowing disorders, rapid motor regression, with limited life expectancy <2 years; type 2 (intermediate) with the onset of symptoms after the age of 6 months, who have acquired the sitting position but will not walk independently. They develop severe, progressive scoliosis from early childhood; type 3 (late) with symptoms after the age of 1 year 6 months, they acquire autonomous gait, but with signs of proximal impairment, motor deficit on the girdles (predominantly on the pelvis), swaying, unstable gait. They do not have bulbar dysfunction, life expectancy is not affected [7,8].

CMAP - Biomarker in SMA

CMAP represents the summation of the motor fibers action potentials in a motor area, it is obtained by the supramaximal stimulation of a peripheral motor nerve [9]. It is an easy-to-obtain but non-specific parameter that does not provide enough information about chronic re-innervation mechanisms. The amplitude of CMAP can be maintained despite the reduction in the number of motor neurons, due to compensatory changes (collateral reinnervation). This aspect is especially encountered in the early stages before a significant loss of the number of motor units [10].

Electrophysiological parameters (CMAP, needle EMG) brought additional information about the cause of motor regression, loss of functional motor neurons, as well as about the compensatory mechanisms of reinnervation [11]. They were initially used as markers of the disease onset, later as follow-up markers in patients treated or not treated with innovative therapies (disease modifiers and gene therapy) [1].

CMAP has also been explored as a prognostic factor. Studies following the natural course of the disease have shown that an initial maximal (baseline) CMAP may have a prognostic role in determining progression in untreated SMA patients. CMAP can be correlated with the pre-existence of an active denervation (degenerative process in progress) and implicitly with the setting of realistic objectives for the therapeutic intervention [12,13].

Patients and Methods

We performed a retrospective study within CNCRNC "Dr Nicolae Robanescu" with patients with SMA type 1, 2, 3 between September 2019 and January 2021.

The inclusion criteria were: 1) pediatric age (0-18 years), 2) genetically confirmed SMA (MLPA) with typical mutation (biallelic mutation in the SMN1 gene) or heterozygous compound, 3) number of SMN2 copies> 2. Exclusion criteria: 1) patients undergoing drug treatment, 2) agitated, uncooperative patients.

CMAP was recorded in the ulnar nerve, by distal supramaximal stimulation. Also, all these patients were assessed from a functional point of view and scores were established on the motor scales corresponding to the type of disease (CHOP for type 1, HFMSE for type 2 and 3). A 6-channel EMG Keypoint device was used for electrophysiological examination. No sedation was administered. A pleasant, comfortable atmosphere was created in the office to ensure the relaxation of the child and of his family in order to have the best possible collaboration [14]. The recorded distal skin temperature was over 35 degrees. We used surface electrodes with adhesive gel (pediatric use) located at the level of the hypothenar eminence - the active electrode (collection) and 3 cm distally from it - the reference electrode. The electrical stimulation was performed at the level of the fist joint, at the level of the ulnar nerve (2-3 cm medially from the median nerve) using, as the case may be, the classic/pediatric stimulator, applying progressive intensities from 10-15 mA to 50 mA. The CMAP obtained was feasible, reproducible, obtained at supramaximal intensities, therefore the electrical stimulation was repeated 3 times. The device software records the maximum amplitude (negative peak).

Needle EMG was performed in all types of SMA (1, 2 and 3) using 0.25x0.35 mm (orange) bipolar needle for pediatric use. Information was collected about spontaneous activity, as well as about the recruitment of motor unit potentials (MUP) under conditions of voluntary contraction. The examination was performed in waking conditions, occasionally using local anesthetic cream. The muscles examined were the anterior tibialis muscle and the brachial biceps.

The functional assessment was performed by a specialized physiotherapist, in good collaboration conditions, with the mother/father, creating a familiar environment. Internationally approved scales were applied for SMA assessment: CHOP INTEND (type 1) and HFMSE (types 2 and 3) [15,16].

CHOP INTEND *scale* [17] is indicated in SMA type 1 and measures the improvement of motor function by assessing active movements, mobility and muscle strength following 16 motor functions grouped into 3 categories: head and neck, hands; arms and shoulders; leg, lower limb and hip. The score varies from 0 to 64.

HFMSE scale [17,18,19] measures motor function in ambulatory to non-ambulatory patients (types 2 and 3). Follows 33 motor functions grouped into 7 categories: sitting position, rolling, transition/crawling, orthostatism/stepping, transition/walking on knees, jumping, stairs. The score can take values between 0-66.

Statistical analysis

For data processing, the Statistical Package for the Social Sciences IBM SPSS Statistics 22 program was used to establish possible correlations between CMAP and scores on functional scales. The tests were considered statistically significant at values of $p < 0.05$ [20].

Results

The group of patients consisted of 36 cases (35 with SMN1 homozygous deletion and 1 case of compound heterozygous) - SMA type 1 - 13 cases, SMA type 2 - 15 cases, SMA type 3 - 8 cases.

From the group of 13 patients with SMA type 1, aged between 3 weeks - 2 years - 92% have 2 SMN2 copies and minimal motor acquisitions: only 38% have head control, rolling 31%, but all require noninvasive ventilation (100%), and on the CHOP scale they obtained scores between 7-59/64 (extremely different, depending on the time interval from the onset of the disease).

Table 1: Clinical signs in SMA

Clinical Signs		Type 1	Type 2		Type 3	
			Sitters	Non-Sitters	Walkers	Non-Walkers
SMN2 copies	2 copy	12 (92%)	3 (20%)	0 (0)	2 (25%)	0 (0)
	3 copy	1 (8%)	11 (73%)	1 (7%)	4 (50%)	2 (25%)
Head controlul		5 (38%)	14 (93%)	1 (7%)	6 (75%)	2 (25%)
rolling		4 (30%)	14 (93%)	1 (7%)	6 (75%)	2 (25%)
Sitting position		0 (0)	14 (93%)	1 (7%)	6 (75%)	2 (25%)
Walking with support		0 (0)	3 (20%)	0 (0)	6 (75%)	0 (0)
Walking without support		0 (0)	0 (0)	0 (0)	4 (50%)	0 (0)
Swallowing disorder		9 (69%)	0 (0)	0 (0)	0 (0)	0 (0)
Noninvasive ventilation		13 (100%)	1 (7%)	0 (0)	0 (0)	0 (0)

Out of 15 patients with SMA type 2 aged 1-14 years - 80% with 3 SMN2 copies, 93% "sitters" with motor acquisitions up to independent sitting, scores with values between 1-36/66 (HFMSE), and 8 patients with SMA type 3 aged 2.6-16 years, 75% with 3 SMN2 copies and 75% ambulatory, with scores of 19-54/66 (HFMSE) (Table 1).

Comparison of CMAP amplitude in patients with SMA versus healthy children of the same age showed significantly lower values in all types of SMA compared to normal (Figure 1)

1. In type 1 the values of the CMAP amplitude were extremely low. Immediately after birth, slightly low values were observed, but after 1-2 weeks, a sudden decrease was registered, values correlated with the motor decline, subsequently remaining constantly low, below 0.5 mV (Figure 2)

Correlations between CMAP amplitude and CHOP scale scores:

Following the comparative statistical analysis between the value of CMAP amplitude and the scores on the CHOP scale, it was observed that there was a significant correlation ($p < 0.03$) between the two parameters, which indicates that patients with SMA type 1 record low CMAP values at CHOP values decrease. (Table 2)

2. In type 2 low values were obtained, sometimes at the lower normal limit, in evolution the values remained relatively constant/stable. Children with type 2a ("weak"/non-sitters) who initially sat, but later lost this ability - showed low values (below 1mV), have amplitudes comparable to type 1, i.e. about 1 mV, and type 2b ("strong"/sitters) with moderately low values, between 2-3 mV (Figure 3).

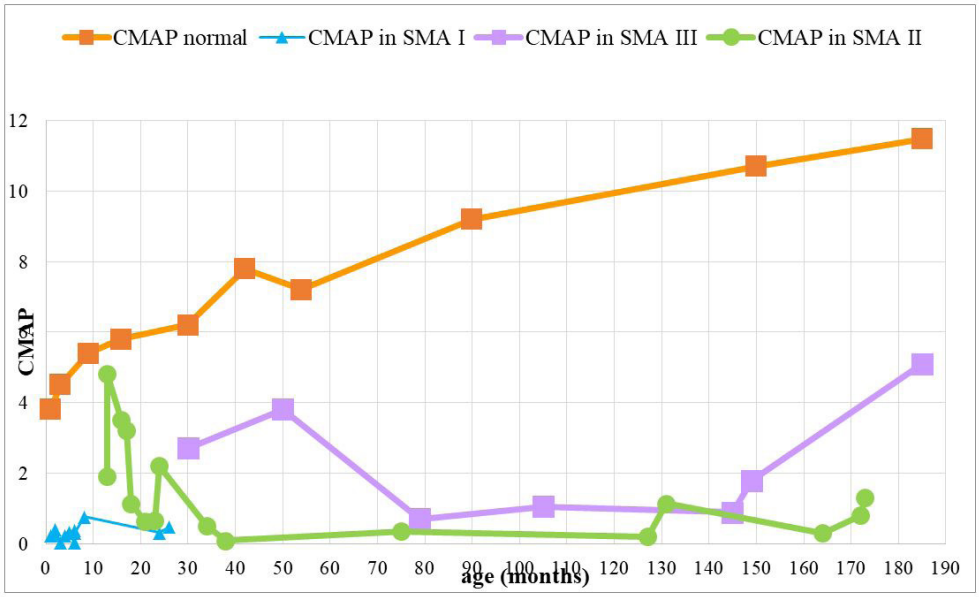

Figure 1: CMAP in SMA type I, II, III vs normal

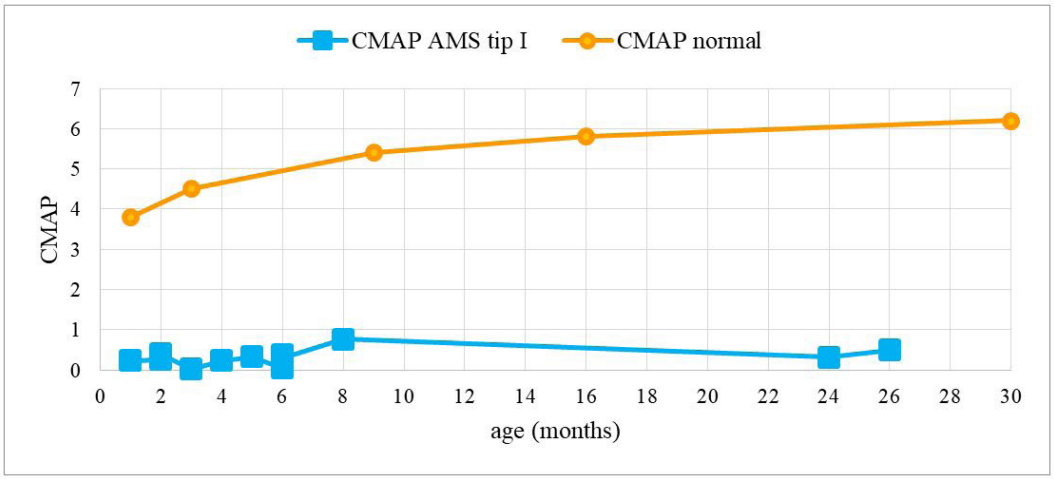

Figure 2: CMAP in SMA type I vs normal

Table 2: CHOP and CMAP1 Values

		CHOP	CMAP1
CHOP	Pearson Correlation	1	*586.
	(Sig. (2-tailed		035.
	N	13	13
CMAP1	Pearson Correlation	*586.	1
	(Sig. (2-tailed	035.	
	N	13	13

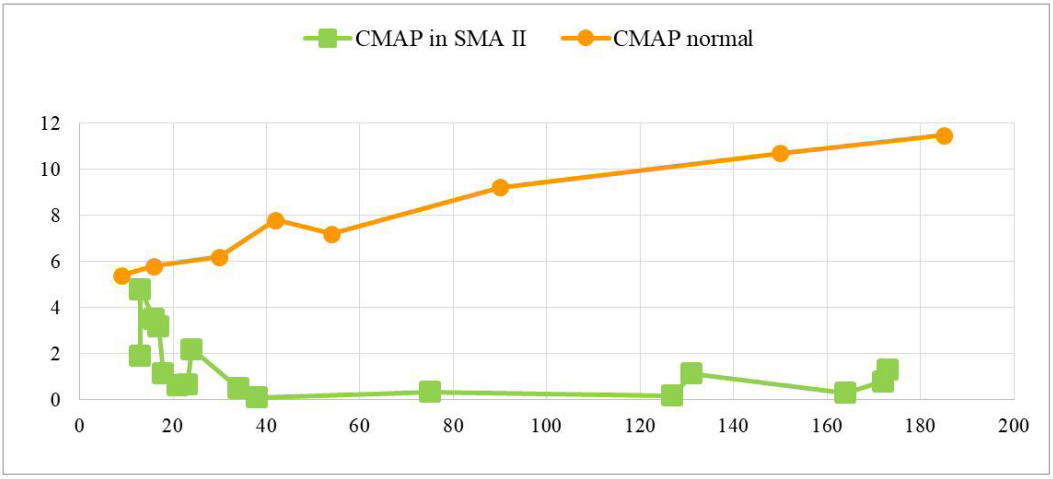

Figure 3: CMAP in AMS tip II vs normal

Correlations between CMAP amplitude and HFMSE scale scores:

A much clearer statistical correlation ($p < 0.005$) was obtained between the CMAP amplitude and the HFMSE scale scores of SMA type 2, indicating a significant concordance between the electrophysiological and the clinical results (Table 3).

3. In type 3 the amplitude of CMAP usually has normal values, registering decreases in the context of motor regression. In the presented group we had patients with form 3a (non-walkers) who lost their autonomic gait relatively fast in evolution (within 2-4 years) who recorded low/very low CMAP amplitude values - approximately 1-2 mV (values that overlap over type 2b). Patients with 3b forms (walkers) who maintained independent gait had almost normal values of CMAP amplitude (Figure 4).

Correlations between CMAP amplitude and HFMSE scale scores:

In type 3 SMA, there was no statistically significant correlation between CMAP amplitude and HFMSE scale scores ($p < 0.1$), indicating a heterogeneous group of patients with inconsistencies between electrophysiological and clinical aspects (Table IV).

Comparing the value of CMAP amplitude between the three SMA groups I, II, III (Figure 10) it was found that they varied depending on the clinical form and patient's age but also on the duration of time from the onset of the disease (motor regression).

Table 3: HFMSE2 and CMAP2 Values

		HFMSE2	CMAP2
HFMSE2	Pearson Correlation	1	**687.
	(Sig. (2-tailed		005.
	N	15	15
CMAP2	Pearson Correlation	**687.	1
	(Sig. (2-tailed	005.	
	N	15	15

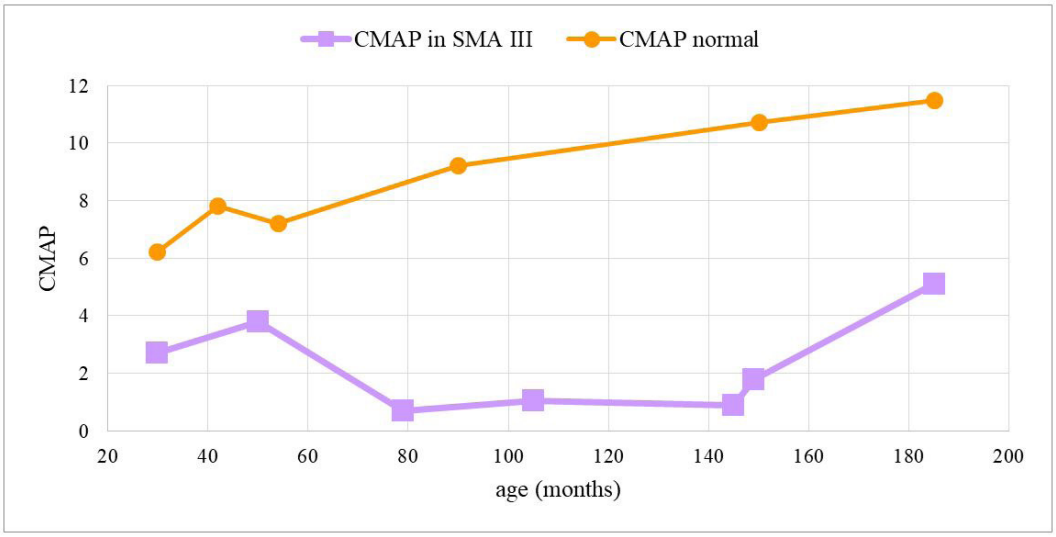

Figure 4: CMAP în AMS tip III vs normal

Table 4: HFMSE2 and CMAP3 Values

		HFMSE3	CMAP3
HFMSE3	Pearson Correlation	1	609.
	(Sig. (2-tailed		109.
	N	8	8
CMAP3	Pearson Correlation	609.	1
	(Sig. (2-tailed	109.	

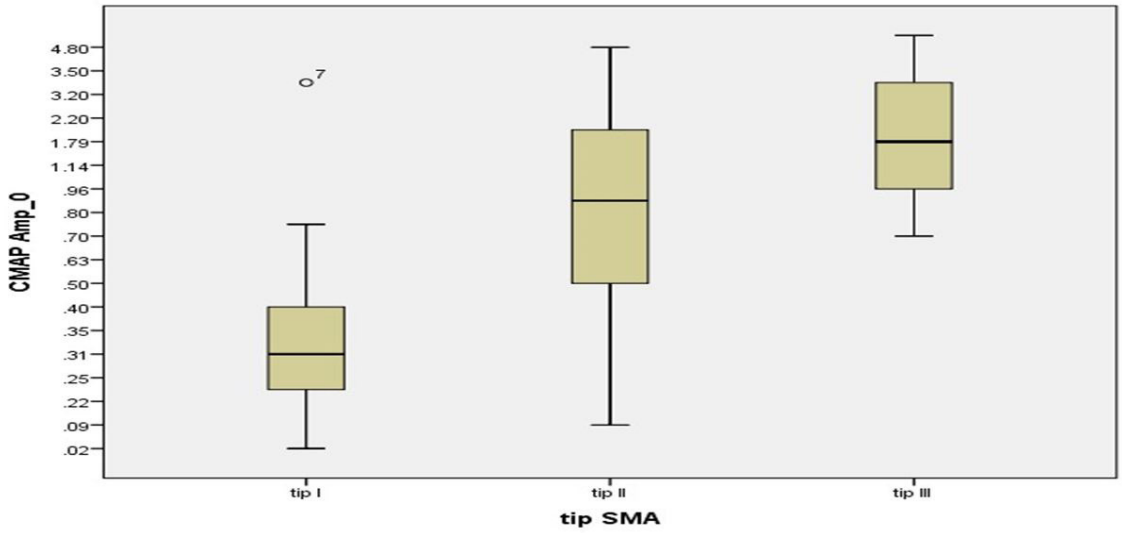

Figure 5: CMAP Amp_0 and tip SMA Values

In the group of patients, we had a *presymptomatic* case aged 1 month (no. 7, Figure 5), without clinical signs of disease (normal muscle tone, osteotendinous reflexes present, without respiratory disorders), with CMAP amplitude values of 3.3 mV (normal for age). He appears graphically in the area of patients with form I but with normal electrophysiological parameters.

Relationship between SMA type and Electromyographic (EMG) Changes

In type 1, the most common EMG aspect was the spontaneous activity present (fibrillation potentials, fasciculations, positive waves) in both the proximal and distal muscles. In type 2 the EMG changes obtained were different depending on the patient's age – 2a ones were similar to those in type 1, but with

the appearance of chronic denervation and re-innervation elements - the recruitment of motor unit potentials (MUP) is reduced, MUP with longer duration in 2b. In the type 3 SMA, the EMG aspect had generally no active denervation elements, with the presence of re-innervation compensating elements (MUP with increased duration and amplitude, late recruitment and incomplete interference). In patients with recent motor regression, it has been correlated with the occurrence of active denervation potentials.

Discussion

The CMAP values in patients with SMA 1, 2, 3 from the analyzed group compared to healthy subjects of the same age was consistent with the data mentioned in the literature. Compared to the normal values of CMAP in healthy subjects [21] who have an average value at the level of the ulnar nerve of 4-11.5 mV (increasing with age), values below this curve [22] (Figure 3) were noticed in all forms of SMA in our group.

The obtained CMAP values raised the suspicion of motoneuron disease, being significantly low in the early forms (1 and 2), and moderately/minimally in the late form [5,11]. These were associated with the time interval from the onset of the disease, being significantly lower in children who already have a significant motor regression and muscle atrophy [23].

Also, in the disease subgroups we observed similar CMAP amplitude values in type 1 and 2a, as well as for 2b and 3a, these were associated with the severity of the clinical picture/motor regression. The electromyographic aspects recorded in our group were similar to those described in the literature [23], with the mention that spontaneous activity with active denervation potentials was directly proportional to the degenerative process/motor regression, and less to the type of disease. In voluntary contraction MUP with modified morphology were registered (increased amplitude and duration, polyphasism), with late recruitment and incomplete interference, these indicating a chronic neurogenic process of re-innervation, more common in late forms.

Similar to the data from the literature [5,8,23,25], an inversely proportional relationship was observed in the analyzed group between the clinical picture and the number of SMN2 copies, the patients with small number of copies having a more severe phenotype of disease. There were 2 exceptions - the presymptomatic case with SMN1 biallelic mutation and 2 SMN2 copies and the one with type 3b - ambulatory, heterozygous compound with 2 SMN2 copies.

Due to the heterogeneous aspect of the examined group and to the relatively small number of examined patients, there were limitations in the statistical analysis. However, it is important to note the importance of associating clinical data with electrophysiological data that gave us a correct perspective on the onset of the disease/motor regression and the possibility of monitoring the natural evolution of the disease versus treatment [12,13,15,18]. Data from the literature have indicated that electrophysiological parameters such as CMAP amplitude associated with clinical ones have been defined in the inclusion criteria in clinical trials such as ENDEAR and SPR1NT [12,26], items monitored in order to follow the evolution of patients undergoing specific treatment and to demonstrate the "amount" of viable motor neurons at the time of treatment initiation for the pre-symptomatic patient.

Also, the value of CMAP amplitude below 1.5 mV in the ulnar nerve and active denervation present (fibrillation, positive waves), along with clinical criteria were proposed as indicators of disease onset in *pre-symptomatic* newborns genetically confirmed with SMA, aspect extremely useful in daily practice because the definition of this category of patients is essential in order to initiate treatment as early as possible [25,27].

Compared to the data from the literature [5,10,24,25,28] and within our group we demonstrated correlations with statistical significance only in the case of type 2/scores on the HFMSE scale ($p < 0.03$, r -0.6) and in the case of type 1/CHOP scores ($p < 0.05$, r-0.5) these demonstrating a synergy between the two parameters at a time of disease evolution. In the case of type 3 of SMA, the lack of statistical significance between CMAP and HFMSE was interpreted in the context of the heterogeneous group of patients, most of them being non-ambulatory patients with long-term chronic evolution.

The limitations of the study consisted in the relatively small number of patients, aged in large intervals within each type of disease, heterogeneous subgroups from the perspective of the time interval between the onset of the disease and the clinical/electrophysiological assessment. Due to these aspects, the statistical significance was not optimal, requiring in the future the expansion of this group of patients, analysis on disease subgroups to ensure better homogeneity.

Also, the evolution of electrophysiological and clinical parameters in patients under new, innovative treatments (disease modifier and gene therapy) will be followed in subsequent studies.

Conclusions

The electrophysiological study was helpful in neuropediatric medical practice along with the clinical examination for raising the suspicion of motoneuron disease (SMA), being a feasible, rapid, easy way to perform investigation. Electrophysiological parameters (CMAP, needle EMG) were used as biomarkers at the onset of the disease, but also in monitoring the evolution, providing significant data for prognosis.

Recognition

The present study is being linked to the whole of the PhD work: "Clinical and electrophysiological correlations in spinal muscular atrophy in patients treated with Nusinersen".

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