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ARTICLE

The relative frequency of common neuromuscular diagnoses in a reference center

Frequência relativa de diagnósticos neuromusculares comuns em um serviço de referência

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ABSTRACT

The diagnostic procedure in neuromuscular patients is complex. Knowledge of the relative frequency of neuromuscular diseases within the investigated population is important to allow the neurologist to perform the most appropriate diagnostic tests. **Objective:** To report the relative frequency of common neuromuscular diagnoses in a reference center. **Methods:** A 17-year chart review of patients with suspicion of myopathy. **Results:** Among 3,412 examinations, 1,603 (46.98%) yielded confirmatory results: 782 (48.78%) underwent molecular studies, and 821 (51.21%) had muscle biopsies. The most frequent diagnoses were: dystrophinopathy 460 (28.70%), mitochondriopathy 330 (20.59%), spinal muscular atrophy 158 (9.86%), limb girdle muscular dystrophy 157 (9.79%), Steinert myotonic dystrophy 138 (8.61%), facioscapulohumeral muscular dystrophy 99 (6.17%), and other diagnoses 261 (16.28%). Conclusion: Using the presently-available diagnostic techniques in this service, a specific limb girdle muscular dystrophy subtype diagnosis was reached in 61% of the patients. A neuromuscular-appropriate diagnosis is important for genetic counseling, rehabilitation orientation, and early treatment of respiratory and cardiac complications.

Keywords: diagnosis; neuromuscular diseases; biopsy; epidemiology.

RESUMO

O procedimento diagnóstico neuromuscular é complexo. O conhecimento da frequência relativa das doenças neuromusculares em uma população é importante para utilização dos testes diagnósticos mais apropriados. **Objetivo:** Relatar a frequência relativa de doenças neuromusculares em um centro de referência. **Métodos:** Revisão de prontuários de pacientes com suspeita de miopatia em 17 anos. **Resultados:** Dentre 3412 exames, 1603 (46,98%) foram confirmatórios: 782 (48,78%) estudos moleculares e 821 (51,21%) biópsias musculares. Os diagnósticos mais frequentes foram: distrofinopatia 460 (28,70%), mitocondriopatia 330 (20.59%), atrofia muscular espinhal 158 (9,86%), distrofia muscular cintura-membros 157 (9,79%), distrofia miotônica de Steinert 138 (8,61%), distrofia muscular face-escápulo-umeral 99 (6,17%) e outros diagnósticos 261 (16,28%). **Conclusão:** Utilizando as técnicas diagnósticas atualmente disponíveis em nosso serviço, o diagnóstico específico do subtipo de distrofia muscular cintura-membros foi obtido em 61% dos pacientes. O diagnóstico neuromuscular apropriado é importante para o aconselhamento genético, orientações de reabilitação e tratamento precoce de complicações respiratórias e cardíacas.

Palavras-chave: diagnóstico; doenças neuromusculares; biópsia; epidemiologia.

The diagnostic investigation of neuromuscular patients is a complex procedure that involves the participation of several professionals. Due to the high diversity of nosologic entities, it is necessary to use different kinds of examinations to confirm the clinical diagnostic hypotheses. It is useful to know the relative frequencies of neuromuscular disorders in the investigated population in order to utilize the most appropriate diagnostic tests. Some neuromuscular reference centers have previously reported on several Brazilian series on the diagnosis of myopathies and these are included on the discussion below[1,2,3,4,5].

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The aim of this study was to describe the relative frequency of neuromuscular diseases in a neuromuscular reference center.

METHODS

This was a retrospective, descriptive study of consecutive molecular examinations and muscle biopsies. Molecular examinations performed between April 19, 1999 and January 18, 2016, and muscle biopsies performed from October 3, 2000 to January 18, 2016 were included.

This study was approved by the Ethics and Research Committee.

Over 17 years, 3,412 patients, admitted to the neuromuscular outpatient clinic, were investigated for diseases in 16 diagnostic categories: 1,200 muscle biopsies and 2,212 molecular exams were performed.

The diagnostic procedure included the correlation between clinical data and laboratory tests, neurophysiology, and image results, with confirmation through molecular examinations, muscle biopsy, and specific enzymatic studies (e.g. alpha-glucosidase) in the patients or their relatives with a confirmed diagnosis. Inclusion criteria were molecular or muscle biopsy diagnoses in 16 categories: 1) Steinert myotonic dystrophy; 2) dystrophinopathy (Duchenne muscular dystrophy, Becker muscular dystrophy, intermediate phenotype dystrophinopathy, and female dystrophinopathy carriers); 3) facioscapulohumeral muscular dystrophy; 4) limb girdle muscular dystrophy, 5) spinal muscular atrophy; 6) congenital muscular dystrophy (except collagen VI disorders); 7) collagen VI neuromuscular disorders (Bethlem myopathy and Ullrich congenital muscular dystrophy); 8) congenital myopathies (subtypes: central core congenital myopathy, nemaline congenital myopathy, centronuclear/myotubular congenital myopathy, cap disease congenital myopathy, and congenital fiber type disproportion); 9) congenital myasthenic syndromes; 10) myofibrillar myopathy; 11) glycogen-storage disease type V (McArdle disease); 12) glycogen-storage disease type II (Pompe disease); 13) dermatomyositis; 14) sporadic inclusion body myositis; 15) other inflammatory myopathies (polymyositis, nonspecific myositis, and necrotizing immune myopathy); and 16) mitochondriopathy.

Exclusion criteria were: 1) myopathic abnormalities without other specifications; 2) neurogenic muscular abnormalities; 3) rare genetic syndromes and other neuromuscular disorders not included in the 16 investigated categories; and 4) duplicated examinations of patients submitted to both muscle biopsy and confirmatory molecular examinations (for duplicated examinations, the results were reported only once on the molecular examination tables) (Table 1).

Table 1. Diagnostic categories: inclusion and exclusion criteria.

Inclusion criteria - 16 diagnostic categories
1) Steinert myotonic dystrophy
2) dystrophinopathy: Duchenne, Becker, intermediate phenotype, and female carriers
3) facioscapulohumeral muscular dystrophy
4) limb girdle muscular dystrophy
5) spinal muscular atrophy
6) congential muscular dystrophy (except collagen VI disorders)
7) Bethlem myopathy/ Ullrich congenital muscular dystrophy (type VI collagenopathy)
8) congenital myopathies (subtypes "central core" congenital myopathy, nemaline congenital myopathy, centronuclear/myotubular congenital myopathy, congenital fiber type disproportion, cap disease congenital myopathy)
9) congenital myasthenic syndrome
10) myofibrillar myopathy
11) glycogen-storage disease type V (McArdle disease)
12) glycogen-storage disease type II (Pompe disease)
13) dermatomyositis
14) sporadic inclusion body myositis
15) other inflammatory myopathies (polymyositis, nonspecific myositis, and immune mediated necrotizing myopathy)
16) mitochondrial myopathy
Exclusion criteria
1) myopathic abnormalities without other specification
2) neurogenic muscular abnormalities
3) rare genetic syndromes and other neuromuscular disorders not included in the 16 inclusion criteria diagnostic categories
4) duplicated diagnosis of patients submitted to both muscle biopsy and confirmatory molecular investigation

Molecular examinations for dystrophinopathies included the investigation of dystrophin gene deletion through polymerase chain reaction (PCR), and deletion and duplication of the dystrophin gene through multiplex ligation-dependent probe amplification.

Molecular investigation of spinal muscular atrophy consisted of the detection of exons 7 and 8 deletion of the survival motor neuron (*SMN)* gene through PCR.

Facioscapulohumeral muscular dystrophy investigation consisted of the search of deletions of repeated tandem 3.3kb units within the D4Z4 locus at the 4q35 region through restriction fragments EcoRI/ HindIII, EcoRI/ AvrII and ApoI through Southern blotting with pulsed field electrophoresis (CHEF-DRIII - Biorad) followed by p13E-11 probe hybridization.

Steinert myotonic dystrophy molecular investigation was performed through the study of the dystrophia myotonica protein kinase (*DMPK*) gene CTG expansion with analysis of EcoRI and BamHI through Southern blotting, followed by pM10M-6 probe hybridization.

Calpainopathy (limb girdle muscular dystrophy type 2A [LGMD2A]) molecular investigation was performed with the search for 24 exons calpain gene point mutations (NM_000070.2) through denaturing high performance liquid chromatography and altered amplicon direct sequencing with the ABI 3130XL Genetic Analyzer (Applied Biosystems Sequencing (v. 5.2) software), and SeqScape (v. 2.5) analysis software, as well as a search for mutation on the Leiden database (www.dmd.nl/index.html).

Sarcoglycanopathy (LGMD2C, LGMD2D, LGMD2E, and LGMD2F) molecular investigation was performed with the study of alpha, beta, gamma, and delta sarcoglycan genes (respectively *SGCD*, *SGCE*, *SGCC*, and *SGCF* genes) through denaturing high performance liquid chromatography followed by altered amplicon direct sequencing with the ABI 3130XL Genetic Analyzer (Applied Biosystems Sequencing (v. 5.2) software), and SeqScape (v. 2.5) analysis software. The exons analyzed were alpha-sarcoglycan exon 3, beta-sarcoglycan exons 3 and 4, delta-sarcoglycan exon 8, and gamma-sarcoglycan exon 6, these being the most frequent mutations in Brazil[6].

Fukutin-related proteinopathy (LGMD2I) molecular investigation was performed with the fukutin-related protein (*FKRP*) investigation through denaturing high performance liquid chromatography followed by altered amplicon direct sequencing with the ABI 3130XL Genetic Analyzer (Applied Biosystems Sequencing (v. 5.2) software), and SeqScape (v. 2.5) analysis software.

The review of muscle biopsy diagnoses was performed through a Systematized Nomenclature of Medicine search in the fields "results" and "observations". The standard procedure for muscle biopsy consisted of histochemical studies and, whenever necessary, immunohistochemical and electron microscopy studies. Histochemical investigations were performed on liquid nitrogen-frozen sections with: hematoxylin and eosin, Gomori modified trichrome, periodic acid-Schiff with and without diastase, Oil-red-O, myosin ATPase (pH9.4, pH4.6, and pH4.3), succinate dehydrogenase, cytochrome-c-oxidase, nicotinamide adenine dinucleotide, acid phosphatase, and non-specific esterase. For patients under investigation for metabolic myopathy, tests for myophosphorylase, phosphofructokinase, and myoadenylate deaminase deficiencies were performed.

Diagnosis of a mitochondrial disorder included definite, probable and possible diagnostic criteria, codified by the Systematized Nomenclature of Medicine, considering histochemical, ultrastructural, molecular (from December 11, 2008), and respiratory chain enzymatic studies (from August 6, 2013).

A dysferlinopathy (LGMD2B) diagnosis was suspected for patients with a characteristic clinical presentation and phenotypic confirmation through immunohistochemistry for dysferlin deficiency.

A congenital myopathy diagnosis followed the International Standard of Care Committee for Congenital Myopathies[7] criteria. Congenital myopathy subtypes included: "central core" congenital myopathy, nemaline congenital myopathy, centronuclear/myotubular congenital myopathy, congenital fiber type disproportion, and cap disease congenital myopathy. A congenital muscular dystrophy diagnosis followed the International Standard of Care Committee for Congenital Muscular Dystrophies criteria[8].

The congenital myasthenic syndrome diagnosis was made by considering anamnesis, clinical examination, and neurophysiological studies with significant repetitive nerve stimulation decrement with or without abnormal ultrastructural neuromuscular junctions[9].

RESULTS

The 2,212 molecular examinations corresponded to diagnostic categories 1 to 5. Category 1: 297 patients with clinical suspicion of Steinert myotonic dystrophy; category 2: 583 patients with clinical suspicion of dystrophinopathy; category 3: 352 patients with clinical suspicion of facioscapulohumeral muscular dystrophy; category 4: 299 patients with clinical suspicion of limb girdle muscular dystrophy, among them 139 patients with clinical suspicion of calpainopathy (LGMD2A), 58 with clinical suspicion of sarcoglycanopathy (LGMD2C/ LGMD2D/ LGMD2E/ LGMD2F), and 102 with clinical suspicion of fukutin-related proteinopathy (LGMD2I); and category 5: 681 patients with clinical suspicion of spinal muscular atrophy.

Of the 1,200 patients submitted to muscle biopsies, 1,199 had consecutive diagnoses with surgical procedures performed between October 3, 2000 and January

18, 2016, and one muscle biopsy had a review of the diagnosis during the study period, based on the muscle biopsy slides on material that had been previously collected on September 21, 1997 (one patient with a diagnosis of Pompe disease).

After the application of inclusion and exclusion criteria (Table 1) to the group of 3,412 patients, 1,603 (47%) patients were selected with confirmed diagnoses in the 16 diagnostic categories, 782 (48.8%) molecular examinations, and 821 (51.2%) muscle biopsies (Figure 1). The most frequent diagnoses were, in descending order: dystrophinopathy 460 (28.70%); definite, probable or possible mitochondriopathy 330 (20.59%); spinal muscular atrophy 158 (9.86%); limb girdle muscular dystrophy 157 (9.79%); Steinert myotonic dystrophy 138 (8.61%); facioscapulohumeral muscular dystrophy 99 (6.17%); congenital muscular dystrophy 79 (4.93%); congenital myopathy 58 (3.62%); polymyositis 35 (2.18%); dermatomyositis 34 (2.12%); sporadic inclusion body myositis 15 (0.94%); type VI collagenopathy 13 (0,81%); myofibrillar myopathy 11 (0.68%); congenital myasthenic syndrome 7 (0.44%); glycogen-storage disease type V/McArdle disease 5 (0.31%), and glycogen-storage disease type II/Pompe disease 4 (0.25%) (Figure 2) (Table 2).

There were 157 patients with limb girdle muscular dystrophy diagnoses. Among them, 34 were excluded from the muscle biopsy diagnosis table because they were submitted to further confirmatory molecular examinations. Therefore, 14 of the 24 patients with calpainopathy (LGMD2A) were excluded from the muscle biopsy table. Of the 26 patients with a sarcoglycanopathy diagnosis, 17 had the diagnosis made through muscle biopsy with immunohistochemistry and nine through molecular examinations; six were excluded from the muscle biopsy table as they had been submitted to both muscle biopsy and molecular examinations. Among the eight patients with a diagnosis of fukutin-related proteinopathy, three were excluded from the muscle biopsy table as they were submitted to molecular examinations after the muscle biopsy. Three patients from the same family received the diagnosis of laminopathy (LGMD1B); three patients from another family received the diagnosis of caveolinopathy (LGMD1C) with the index case submitted to muscle biopsy, and three patients received the diagnosis of telethoninopathy (LGMD2G), confirmed through muscle biopsy with immunohistochemical deficiency on telethonin expression. With the presently-available techniques in our service, a confirmatory diagnosis of limb girdle muscular dystrophy subtype was made in 61% of the patients (Table 3)[5,10,11,12,13,14].

There were 344 male patients with a diagnosis of dystrophinopathy. Among the 287 patients with a diagnosis of Duchenne muscular dystrophy, 184 were confirmed by molecular examination and 103 by muscle biopsy. Among 45 patients with Becker muscular dystrophy, 33 were confirmed by molecular examination and 12 by muscle biopsy. Among the 12 patients with intermediate dystrophinopathy phenotype, 10 were confirmed by molecular examination and two by muscle biopsy; four patients were excluded from the muscle biopsy table as they were later submitted to confirmatory molecular examination.

The initial date of the examinations varied according to the specific technique implementation: Steinert myotonic dystrophy molecular examinations started on April 19, 1999, dystrophinopathy on June 4, 2001, spinal muscular atrophy on May 29, 2000, facioscapulohumeral muscular dystrophy on February 12, 2003, calpainopathy (LGMD2A) on May 24, 2011, sarcoglycanopathy (LGMD2C, LGMD2D,

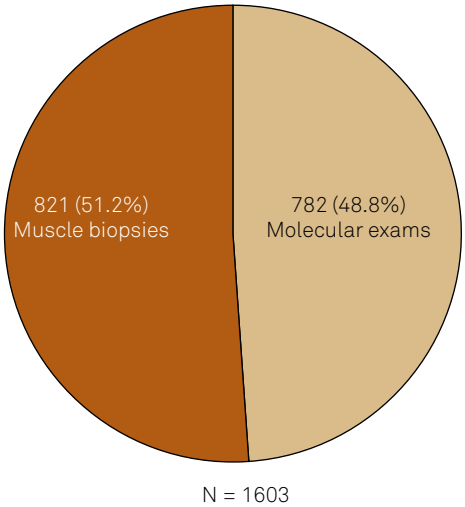

Figure 1. Muscle biopsy-molecular neuromuscular examination ratio.

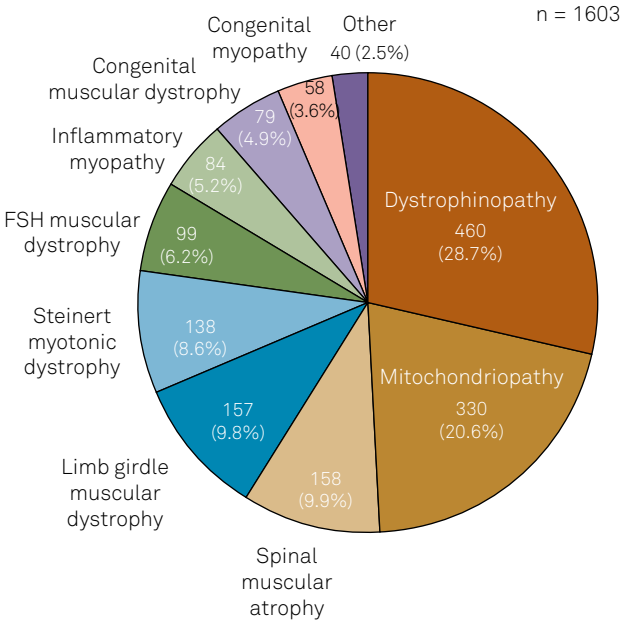

Figure 2. Relative frequencies of neuromuscular diagnoses.

LGMD2E, and LGMD2F) on May 26, 2011, fukutin-related proteinopathy on May 18, 2011. Muscle biopsies started in 1997 and the informatized diagnostic system, codified through the Systematized Nomenclature of Medicine, started on October 3, 2000.

After April 5, 2013, the diagnosis of dysferlinopathy was made through muscle biopsy with immunohistochemical deficiency in dysferlin expression after the confirmation of normal caveolin immunohistochemical expression and absent calpain mutation[15,16,17].

Table 2. Relative frequency of neuromuscular disorders in a reference center.

Neuromuscular disorders	Number of patients		Subtypes	
	n	%	n	% within category
Steinert myotonic dystrophy	138	8.61		
Mitochondriopathy	330	20.59		
Dystrophinopathy - total of male patients	344	21.46		
Duchenne type dystrophinopathy			287	83.43[a]
Becker type dystrophinopathy			45	13.08[a]
Intermediate phenotype dystrophinopathy			12	3.49[a]
Asymptomatic female dystrophinopathy carriers	104	6.49		
Symptomatic female dystrophinopathy carriers	12	0.75		
Facioscapulohumeral muscular dystrophy	99	6.17		
Limb girdle muscular dystrophy - total	157	9.79		
Laminopathy (LGMD1B)			3	1.91[b]
Caveolinopathy (LGMD1C)			3	1.91[b]
Calpainopathy (LGMD2A)			24	15.29[b]
Dysferlinopathy (LGMD2B)			29	18.47[b]
Sarcoglycanopathy (LGMD2C, 2D, 2E, 2F)			26	16.56[b]
Fukutin-related proteinopathy (LGMD2I)			8	5.10[b]
Telethoninopathy (LGMD2G)			3	1.91[b]
Undetermined			61	38.85[b]
Spinal muscular atrophy	158	9.86		
Congenital muscular dystrophy - total	79	4.93		
Merosin negative			16	20.25[c]
Type VI collagenopathy (Bethlem/ Ullrich)	13	0.81		
Congenital myopathies - 5 subtypes	58	3.62		
Central core/ multiminicore			26	44.83[d]
Nemaline			16	27.59[d]
Centronuclear/myotubular			11	18.97[d]
Cap disease			4	6.89[d]
Congenital fiber type disproportion			1	1.72[d]
Congenital myasthenic syndrome	7	0.44		
Myofibrillar myopathy	11	0.68		
Glycogenosis type V (McArdle disease)	5	0.31		
Glycogenosis type II (Pompe disease)	4	0.25		
Inflammatory myopathies - total	84	5.24		
Dermatomyositis			34	40.47[e]
Sporadic inclusion body myositis			15	17.86[e]
Polymyositis/nonspecific myositis/ immune-mediated necrotizing myopathy			35	41.67[e]
Total	1603	100		

a: percentage from the total male dystrophinopathy patients; b: percentage of limb girdle muscular dystrophy; c: percentage of congenital muscular dystrophy; d: percentage of the five most common congenital myopathy subtypes; e: percentage of inflammatory myopathy.

Table 3. Relative frequency of limb girdle muscular dystrophies.

Subtype of limb girdle muscular dystrophy	Present study		Brazil		UK		Italy		Italy		Mexico		Australia	
			Zatz et al., 2003[5]		Norwood et al., 2009[14]		Fanin et al., 2009[10]		Guglieri et al., 2008[11]		Gómez-Díaz et al., 2012[12]		Lo et al., 2008[13]	
	n = 157 patients		n = 120 families		n = 43 patients		n = 346 patients		n = 155 families		n = 97 patients[b]		n = 76 patients	
	n	%	n	%	n	%	n	%	n	%	n	%	n	%
LGMD1B laminopathy	3	2	NR	NR	6	14	5	2	NR	NR	NR	NR	1	1
LGMD1C caveolinopathy	3	2	NR	NR	NR	NR	5	2	2	1	3	3	2	3
LGMD2A calpainopathy	24	15	38[a]	32	15	35	87	25	44	28	24	25	6	8
LGMD2B dysferlinopathy	29	18	27[a]	22	2	5	39	11	29	19	39	40	4	5
LGMD2C-2D-2E-2F sarcoglycanopathy	26	17	38[a]	32	8	18	52	15	28	18	30	31	2	2
LGMD2I fukutin-related proteinopathy	8	5	13[a]	11	12	28	15	4	10	7	NR	NR	2	3
LGMD2G telethoninopathy	3	2	4[a]	3	NR	NR	NR	NR	NR	NR	NR	NR	NR	NR
Indeterminate	61	39	NR	NR	NR	NR	141	40	42	27	NR	NR	59	78[c]
Other dystrophies	NR	NR	NR	NR	NR	NR	2	1	NR	NR	1	1	NR	NR

NR: not reported; a: absolute number of patients calculated based on the published percentages; b: initial number of 290 biopsies (dystrophin and merosin deficiencies were excluded); c: 78 indeterminate, of whom 58 have normal dysferlin expression and 20 have defective dysferlin. The frequencies of sarcoglycanopathy were calculated with the total of patients of each subtype: Fanin 2009[10], LGMD2C-2F = 11+30+10+1 = 52 (15%); Guglieri 2008[11], LGMD2C-2F = 32 = 4.5+8.4+4.5+0.7=18.1%. LGMD1B = laminopathy (limb girdle muscular dystrophy type 1B – *LMNA* gene mutation), LGMD1C = caveolinopathy (limb girdle muscular dystrophy type 1C – *CAV3* gene mutation); LGMD2A = calpainopathy (limb girdle muscular dystrophy type 2A, *CAPN3* gene mutation), LGMD2B = dysferlinopathy (limb girdle muscular dystrophy type 2B, *DYSF* gene mutation), LGMD2C-2D-2E-2F = sarcoglycanopathy (limb girdle muscular dystrophy types 2C-2D-2E-2F, LGMD2C gamma-sarcoglycan (*SGCG*) gene mutation, LGMD2D alpha-sarcoglycan (*SGCA*) gene mutation, LGMD2E beta-sarcoglycan (*SGCB*) gene mutation, and LGMD2F delta-sarcoglycan (*SGCD*) gene mutation; LGMD2I = fukutin-related proteinopathy (limb girdle muscular dystrophy type 2I, *FKRP* gene mutation) LGMD2G = telethoninopathy (limb girdle muscular dystrophy type 2G, *TCAP* gene mutation).

DISCUSSION

This study provided an estimated relative frequency of specific diagnoses (16 diagnostic categories) through molecular and muscle biopsy examinations in a neuromuscular reference clinic, over the last 17 years. About half the patients received a diagnosis, within one of the 16 categories, by molecular studies, and half by muscle biopsy. The relative frequency of neuromuscular diseases showed some similarities to the neuromuscular disease prevalence in a study in northern England[14]. In that study, the most frequent diagnoses, in descending order, were: myotonic dystrophy, mitochondriopathy, dystrophinopathy, facioscapulohumeral muscular dystrophy, limb girdle muscular dystrophy, spinal muscular atrophy, congenital muscular dystrophy, Bethlem type VI collagenopathy, and congenital myopathies (central core and nemaline)[14]. On the other hand, in the present study, the most frequent diagnoses, in descending order were: dystrophinopathy, mitochondriopathy, spinal muscular atrophy, limb girdle muscular dystrophy, Steinert myotonic dystrophy, and facioscapulohumeral muscular dystrophy. Due to methodological differences between both studies, it is not possible to conclude that the different relative frequencies of myotonic dystrophy in both studies should be attributed to differences in prevalences. Besides, only patients with myotonic dystrophy type 1 (Steinert myotonic dystrophy) were included in the present study, and type 2 myotonic dystrophy patients were not included in our study.

One limitation of this study is related to the diagnosis of dysferlinopathy, as the diagnosis was immunophenotypical and not genotypical. Considering the possibility of immunohistochemical secondary deficiencies, after April 5, 2013, the diagnosis was made taking into consideration the absence of calpain gene mutations, detection of calpain expression on Western blot, and detection of immunophenotypical caveolin expression[15,16,17] (Table 3)[5,10,11,12,13,14].

Considering the diagnostic techniques available over the last 17 years, a conclusive, definite limb girdle muscular dystrophy subtype diagnosis was achieved in about 61% of the patients. In the previously-mentioned study in northern England, using muscle biopsy, immunohistochemistry, Western blot, and genetic sequencing, a conclusive limb girdle muscular dystrophy subtype diagnosis was achieved in 75% of the patients[18], whereas in another study in Italy, the percentage of confirmed limb girdle muscular dystrophy subtype diagnoses was 60%[10,18].

Even after the utilization of molecular examinations during the diagnostic procedure at the neuromuscular outpatient clinic, muscle biopsies represented 51.2% (821/1,603) of the examinations used to confirm the diagnosis. In a French study, muscle biopsies resulted in 43.6% of conclusive diagnoses[19]. In that study, the most frequent muscle biopsy diagnoses, in descending order were: congenital myopathy (for the most part corresponding to nonstructural congenital myopathy), progressive muscular dystrophy (for the most part corresponding to dystrophinopathy), mitochondrial encephalomyopathy, other metabolic myopathies, congenital muscular dystrophy,

and dermatomyositis[19]. In the present study, the most frequent muscle biopsy diagnoses were, in descending order: mitochondriopathy, dystrophinopathy, limb girdle muscular dystrophy, inflammatory myopathy (polymyositis, dermatomyositis, and others), congenital muscular dystrophy, and congenital myopathy. The difference in the frequency of congenital myopathy between these studies may be due to the inclusion criteria as, in the present study, only five structural congenital myopathy subtypes were included (central core congenital myopathy, nemaline congenital myopathy, centronuclear/myotubular congenital myopathy, congenital fiber type disproportion, and cap disease congenital myopathy).

An analysis of 4,500 muscle biopsies performed from 1979 to 2012 in another Brazilian reference center identified 19 patients with Pompe disease[1]; the same reference center reported 106 patients submitted to dystrophin gene DNA analysis between 1999 and 2005 and at least one deletion was detected in 76 cases[2], as well as 56 patients with limb girdle muscular dystrophy submitted to muscle biopsy from 1976 to 2001[3]. From a group of 3,802 patients submitted to muscle biopsy between 1989 and 2001, at another Brazilian neuromuscular reference center, 86 patients were found to have mitochondriopathy of the chronic progressive external ophthalmoplegia subtype[4]. Yet another Brazilian group reported 120 unrelated families with limb girdle muscular dystrophy, submitted to molecular investigation until 2003[5].

With the advent of novel technologies, such as next generation sequencing, there is hope that the resolution rate of molecular studies may increase with consequent reduction in the number of undetermined diagnoses. This would provide the patients with: 1) adequate genetic counseling; 2) early clinical, respiratory and cardiac management; 3) rehabilitation orientation concerning gait prognosis and daily activities, according to the subtype of neuromuscular disorder.

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