Expanding the Phenotypic and Genetic Spectrum of Neuromuscular Diseases Caused by DYNC1H1 Mutations

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Objectives: Spinal muscular atrophy with lower extremity predominance 1 (SMALED1) and Charcot–Marie-Tooth disease type 2O (CMT2O) are two kinds of hereditary neuromuscular diseases caused by DYNC1H1 mutations. In this study, we reported two patients with SMALED1 caused by DYNC1H1 mutations. The genotype–phenotype correlations were further analyzed by systematically reviewing previous relevant publications.

Materials and Methods: Two patients’ with SMALED1 and their parents’ clinical data were collected, and detailed clinical examinations were performed. WES was then applied, which was confirmed by Sanger sequencing. PubMed, Web of Science, CNKI, and Wanfang Data were searched, and all publications that met the inclusion criteria were carefully screened. Any individual patient without a detailed description of clinical phenotypes was excluded.

Results: The two patients manifested delayed motor milestones and muscle wasting of both lower extremities. The diagnosis was further confirmed as SMALED1. Genetic testing revealed heterozygous DYNC1H1 mutations c.1792C>T and c.790C>G; the latter is a novel dominant mutation. Genotype–phenotype analysis of DYNC1H1 variants and neuromuscular diseases revealed that mutations in the DYN1 region of DYNC1H1 protein were associated with a more severe phenotype, more complicated symptoms, and more CNS involvement than the DHC_N1 region.

Conclusion: Our study potentially expanded the knowledge of the phenotypic and genetic spectrum of neuromuscular diseases caused by DYNC1H1 mutations. The genotype–phenotype correlation may reflect the pathogenesis underlying the dyneinopathy caused by DYNC1H1 mutations.

Keywords: neuromuscular disease, spinal muscular atrophy, Charcot-Marie-Tooth (CMT) disease, DYNC1H1, genotype-phenotype correlation
INTRODUCTION

Located on 14q32.31, the DYNC1H1 gene encodes dynein cytoplasmic 1 heavy chain 1 (DYNC1H1), which is the core structure of cytoplasmic dynein. Dynein is a large (~1.5 MDa) motor protein complex responsible for retrograde axonal transport in all eukaryotic cells (1). DYNC1H1 plays an essential role in ATPase-dependent movement along the microtubule and recruitment of other dynein subunits (2). Mutations in DYNC1H1 can lead to various developmental and degenerative diseases of the nervous system, which are nominated as a disease spectrum of “dyneinopathy” characterized by locomotor and motor system deficits, sensory system defects, and (or) brain morphology and function abnormalities (3).

Spinal muscular atrophy with lower extremity predominance (SMALED1, OMIM: 158600) and Charcot–Marie type 20 (CMT2O, OMIM: 614228) have both been reported as hereditary neuromuscular diseases caused by DYNC1H1 variants (4–6). Spinal muscular atrophy (SMA) is caused by the impairment of motor neurons in the spinal cord. SMALED1 is an autosomal dominant hereditary type of SMA, characterized by non-length-dependent weakness restricted or predominant in the lower limbs (4, 7, 8). Charcot–Marie-tooth (CMT) disease, also known as hereditary motor and sensory neuropathy (HMSN), is genetically heterogeneous and clinically characterized by progressive distal muscle weakness and wasting, sometimes accompanied by sensory abnormalities. CMT2O was first reported by Weedon et al. in a large four-generation family and the patients presented with delayed motor milestones, abnormal gait, and slowly progressive distal lower limb atrophy and weakness accompanied by pes cavus deformity (6).

Although numerous cases have been reported worldwide, the analysis of the genotype–phenotype correlations between DYNC1H1 mutations and neuromuscular diseases, including SMALED1 and CMT2O, is still scarce. Here, we first reported DYNC1H1 gene c.1792C>T (p.R598C) and c.790C>G (p.R264G) de novo heterozygous mutations in two sporadic SMALED1 cases. Next, we reviewed previous publications and summarized the phenotypic and genetic characteristics of neuromuscular diseases caused by DYNC1H1 mutations. Furthermore, we focused on the genotype–phenotype correlations of DYNC1H1 variants, which may provide insights into unraveling the mechanism of the clinical heterogeneity of “dyneinopathy.”

METHODS

Ethics

The study was approved by the Ethics Committee of Huashan Hospital, Fudan University. The patients and their family members all provided informed consent according to the Declaration of Helsinki.

Whole-Exome Sequencing (WES) and Candidate Variant Screening

Genomic DNA was extracted from EDTA-treated peripheral blood using a DNA extraction kit (Qiagen, Hilden, Germany). The purity and quality of DNA were examined by 1% agarose electrophoresis and Qubit, respectively. The DNA was then sequenced by WES. All exons of the two probands were sequenced by the HiSeq4000 Illumina Genome Analyzer II platform (Illumina, San Diego, CA). The variants were analyzed according to the previous reference (9): (1) automatically including variants with minor allele frequencies ≤0.01 in further analysis by sifting of variants against the Human Genome Mutation Database (HGMD) (http://www.hgmd.cf.ac.uk/ac/index.php), the NCBI SNP database (https://www.ncbi.nlm.nih.gov/snp), and the Exome Variant Server (10); (2) filtering those variants which do not code protein and do not alter splice sites as predicted by BDGP (https://www.fruitfly.org/seq_tools/splice.html); (3) ignoring synonymous variants; and (4) combining clinical manifestations and genetic characteristics to screen for associated variants. The detected DYNC1H1 variants were checked or adjusted to ensure that they matched with the reference transcript NM_001376.5 for DYNC1H1.

Analysis and Interpretation of DYNC1H1 Variants

Sanger sequencing was performed to confirm the mutations and co-segregations within family members. Various in silico algorithms were applied to interpret the pathogenicity of detected variants, including MutationTaster (http://www.mutationtaster.org/), PolyPhen-2 (http://genetics.bwh.harvard.edu/pph2/), SIFT (http://provean.jcvi.org/ genome_submit_2.php?species=human), and PROVEAN (http://provean.jcvi.org/ genome_submit_2.php?species=human). The pathogenicity of gene variations was classified according to the classification standard of the American College of Medical Genomics (ACMG) (10). Protein sequences across species were aligned using the ClustalW algorithm via MEGA11 software.

Collection of Previous Publications

Online databases, including PubMed, Web of Science, CNKI, and Wanfang Data, were searched, applying the following search terms from January 1980 to April 2021: (mutation OR variant) AND [DYNC1H1 AND ("Charcot-Marie-Tooth Disease" OR “CMT” OR “hereditary motor and sensory neuropathy” OR “HMSN” OR “spinal muscular atrophy” OR “SMA”)]. Human Gene Mutation Database (HGMD) (http://www.hgmd.cf.ac.uk/), Online Mendelian Inheritance in Man (OMIM) (http://www.omim.org/), and Ensembl (http://uswest.ensembl.org/index.html) were also searched with the search term DYNC1H1. All resulting publications were carefully screened. We excluded publications that (1) were reviews, (2) were laboratory studies, (3) were not related to neuromuscular diseases, and (4) did not involve any DYNC1H1 variants. Among the included publications, families or a single patient with no detailed description of clinical phenotypes were also excluded from this study to improve the quality of analysis. Clinical and genetic data were collected from each reported patient. All the variants were checked or adjusted to ensure that they matched with the reference transcript NM_001376.5 for DYNC1H1. All the processes were performed independently by two authors (J.L.)
and S.D.), and any discrepancy in the assessment were resolved by consensus.

**Statistics**

The distributions of continuous variables were tested for normality using the Kolmogorov–Smirnov test, and the difference between the two groups was compared using Student's t-test or the Mann–Whitney test, wherever appropriate. The chi-squared test or the Fisher exact test was used to compare the categorical variables. Moreover, Bonferroni adjustment was also applied in multiple comparisons. All analyses were two-tailed, and a p-value < 0.05 was considered statistically significant. All the statistical data were analyzed using SPSS 20.0 (SPSS Inc., Chicago, IL, USA).

**RESULTS**

**Clinical and Genetic Analysis of Two SMALED1 Cases**

Case 1 is a 14-year-old female patient who complained of difficulty standing upright from a squatting position for over 1 year. The patient learned to walk at 14 months of age and ran slower than her peers since childhood. On physical examination, the patient presented with bilateral pes cavus and muscle wasting of distal lower limbs. Muscle strength was normal, except for 4/5 weakness of bilateral distal upper limbs and lower limbs. Tendon reflexes were attenuated in all limbs, and Babinski's sign was negative. The sensory system was normal on examination. The patient also showed an unaffected intelligence level. Her parents had normal manifestations, and there was no related family history (Figure 1A).

Case 2 is a 15-year-old male patient who sought clinical help for difficulty standing upright from a squatting position for over 1 year. The patient could walk since 19 months of age and had been prone to fall since childhood. Physical examination showed mild lordosis and muscle wasting of both lower limbs (Figure 1C). Muscle strength was normal, except for 4/5 weakness of bilateral distal lower limbs. Tendon reflexes were attenuated. Babinski's sign was absent. The patient's sensory examination was normal with an unaffected intelligence level. His parents were both normal, and no related family history was declared (Figure 1B).

Laboratory examinations revealed normal (181 U/L) and elevated (294 U/L) levels of serum creatine kinase (CK) in the two patients, respectively. In case 1, electromyogram (EMG) detected no fibrillation potential or positive sharp wave in the right big toe, the tibialis anterior, and the rectus abdominis. The affected amino acids were conserved across vertebrates (Figure 1G). Various bioinformatics software packages (Mutation Taster, Polyphen2, SIFT, and PROVEAN) were applied to examine the pathogenicity of the two mutations. Both mutations were predicted as “pathogenic” according to ACMG guidelines.

**DYNC1H1 Mutations in Neuromuscular Diseases**

Previous publications were searched to further explore the clinical and genetic spectrums of DYN1H1-related neuromuscular diseases. A total of 22 original articles out of 49 publications met the inclusion criteria and were included in our study (Supplementary Figure 1). These articles identified 39 distinct variants in DYN1H1 related to neuromuscular diseases. Of the 105 patients with DYN1H1 mutations reported by these publications, SMALED1 (77, 73.3%) and CMT20 (16, 15.2%) accounted for the most part. Among the patients with SMALED1, two were initially diagnosed with polio and chronic inflammatory demyelinating polyradiculoneuropathy (CIDP). Other types of neuromuscular diseases, such as hereditary spastic paraplegia (HSP), were also reported to be caused by DYN1H1 variants. Of all the 58 families reported, most were European families (34/58, 58.6%), followed by East Asia (9/58, 15.5%).
FIGURE 1 | Genetic analysis and clinical phenotypes of two SMALED1 pedigrees. (A) Family tree of patient 1. (B) Family tree of patient 2. (C) Patient 2 presented with obvious muscle wasting in both distal lower limbs. (D) Sanger sequencing of Patient 1 and her parents revealed a de novo DYNC1H1 gene mutation c.1792C>T. (E) Sanger sequencing of patient 2 and his parents revealed a de novo DYNC1H1 gene mutation c.790C>G; the affected member is marked in black, and the proband is indicated by arrows. (F) Affected amino acids R598 and R264 were conserved in human and various other vertebrates. (G) Pathogenicity prediction of the mutations by several in silico bioinformatic tools and according to ACMG guidelines.
North America (8/58, 13.8%), Australia (5/58, 8.6%), and the Middle East (2/58, 3.4%).

Mutational Spectrum of DYNC1H1 Associated With Neuromuscular Diseases

All variants were missense mutations, except for one splicing variant (c.12685-3C>T); two patients from a Chinese family had complex mutations in DYNC1H1 (c.2419G>A and c.12685-3C>T). To analyze the exon distribution of the missense variants, the average frequency of variants per length was calculated to balance the length of exon variation. Analysis of DYNC1H1 variants revealed that exon 8 carried the most amount of DYNC1H1 variants (Figure 2A). However, after adjusting for exon length, we found that exons 4, 14, 5, and 8 were the top four hot regions related to neuromuscular diseases, with variants per length estimated to be 0.0273, 0.0270, 0.0267, and 0.0223, respectively (Figure 2B). Several recurrent DYNC1H1 variants were reported, with c.1792C>T (p.R598C) being the most common mutation (7/58 families with 10 patients, 12.1%). Others were c.751C>T (6/58, 10.3%), c.1953G>A (4/58 6.9%), c.2327C>T (3/58, 5.2%), c.917A>G (3/58, 5.2%), and c.3170A>G (2/58, 3.4%). Interestingly, although c.1792C>T (p.R598C) was suggested to be the most common mutation site related to neuromuscular diseases worldwide, it has not been reported in the East Asian population in previous publications. Instead, c.751C>T (3/9 families, 33.3%) and c.2327C>T (2/9 families, 22.2%) were more commonly reported in this area.

Human DYNC1H1 protein is composed of 4646 amino acids (aa). It can be roughly divided into the tail region (0-1450 aa) and the motor region (1450-4646 aa) according to its structural and functional characteristics (11). Despite that the motor region was longer, 27 of the 39 variants (69.2%) were distributed in the tail region, and only 12 of the 39 (30.8%) were located in the motor region, which was in correspondence with previous studies that mutations in the tail region were more commonly related to motor-related disorders (4, 12). More precisely, the DYNC1H1 protein is divided into several domains overlapping each other (Figure 2C). Cases involved in our study revealed that the stem domain (53-1867 aa) contained the most amount of DYNC1H1 mutations (30/39, 76.9%), among which 18 of the 39 (46.2%) were located in overlapping regions of the stem with other domains, including DHC_N1 (242-832 aa), dynein intermediate chain (DIC)-binding domain (448-703 aa), and dynein intermediate light chain (DILC)-binding domain (651-802 aa). Mutations in other domains were relatively less frequent, including three ATPase families associated with various cellular activity (AAA) domains (2180-3168 aa, 7/39, 17.9%) and stalk (3189-3500 aa, 2/39, 5.1%) (Figure 2C). This result may be due to the fact that the stem is the longest domain of DYNC1H1 located more closely to the N terminal and overlaps with many other regions. The detailed information on the DYNC1H1 genetic spectrum of patients with neuromuscular diseases is listed in Supplementary Table 1.

Apart from neuromuscular diseases such as SMALED1 and CMT2O, DYNC1H1 gene mutation is also related to intellectual disability characterized by early-onset seizures, mild dysmorphic features, and cortical malformations (13-15). CMT2O is uniquely linked with DYNC1H1 gene mutations, while SMALED has also been reported to be associated with BICD cargo adaptor 2 (BICD2) mutations (8, 16). BICD2 protein is important in dynein complex binding, and animal models indicated that BicD2 knockdown mouse embryos showed significant inhibition of neuronal migration (8, 17). The relationship between SMALED, CMT2O, and DYNC1H1 mutations is illustrated in Figure 2D.

Clinical Spectrum of Neuromuscular Diseases Caused by DYNC1H1 Mutations

A total of 105 patients with neuromuscular diseases were reported carrying DYNC1H1 mutations. Of 66 patients with exact ages provided, the average age was 21.8±19.7 (range, 1.0-82.0) years. The onset age of the patients with DYNC1H1 mutations was relatively young, with more than two-thirds of the patients showing disease onset at birth or in infancy (<1 year old) (Figure 2E). The detailed clinical information of patients with neuromuscular diseases caused by DYNC1H1 mutations is listed in Supplementary Table 2. Muscle weakness of lower limbs was the most common clinical phenotype of these patients, among which nearly two-thirds presented with proximal dominant weakness of lower limbs. In comparison, more patients showed distal dominant atrophy than proximal dominant atrophy. This may be due to the larger volume of proximal muscles in the lower limbs, which can hide the relatively small degree of proximal muscle wasting. A small part of the patients also had upper limb involvement. Delayed motor milestones were usually the earliest clinical manifestation of patients with SMALED1 or CMT2O and were reported in more than half of the patients. However, most patients remained ambulant, and Gowers sign was not common, suggesting the relatively mild degree of lower limb impairment and the rare involvement of truncal muscle in these groups of diseases. The sensory system was usually exempted from the diseases, and the involvement of the central nervous system (CNS) was also relatively less common, with around one-third of patients having intellectual disability and around one-tenth of them experiencing seizure episodes. Most patients showed deformities predominantly involving the musculoskeletal system, of which pes cavus and joint contracture or deformities were the most common. Other deformities, including pes planus, hand deformities, spine deformities, scapular winging, exotropia, and aortic root dilation were also observed in a number of patients. The percentage of some clinical phenotypes is shown in Figure 2F.

Decreased or absent tendon reflex was the most notable clinical phenotype on physical examination. The serum CK level and NCV test showed abnormal results only in a small percentage of patients. Abnormal brain MRI was more common and was detected in about 60% of patients.

Genotype–Phenotype Correlations of DYNC1H1 Variants

To further explore the phenotypic and genetic spectrum of DYNC1H1 mutations, we then analyzed the genotype–phenotype correlations between DYNC1H1 variants and neuromuscular
FIGURE 2 | Genetic and phenotypic spectrum of neuromuscular diseases caused by DYNC1H1 mutations. (A) DYNC1H1 variants on different exons not adjusting for exon length. (B) DYNC1H1 variants on different exons adjusting for exon length by calculating variant amounts per length. (C) Structural model of DYNC1H1 protein and relevant mutation sites in previous publications associated with neuromuscular diseases; the numbers in brackets indicate the recurrent times of mutations in different pedigrees; (D) Relationship between SMALED1, CMT2O, and DYNC1H1 mutations. (E) Distribution of disease onset age of patients with neuromuscular diseases caused by DYNC1H1 mutations. (F) Radar chart shows the percentage of some common features of DYNC1H1-related neuromuscular diseases. AAA, ATPase family associated with various cellular activities; ARX, aristaless-related homeobox; BICD2, BICD cargo adaptor 2; CMT2O, Charcot–Marie tooth type 2O; DIC, dynein intermediate chain; DILC, dynein intermediate light chain; DLL, distal lower limbs; FRMPD4, FERM and PDZ domain containing 4; LL, lower limbs; PLL, proximal lower limbs; SMALED, spinal muscular atrophy with lower extremity predominance; UL, upper limbs.
Dysfunctional DYNC1H1 mutations leading to neuromuscular diseases in the current study, none were located in Dynein_C. Therefore, the DYNC1H1 mutations affecting the DHC/N1 domain may have a more severe phenotype, more complicated clinical manifestations, and more CNS involvement than those who had mutations in the DHC/N1 region.

Covering a length of 19940 base pairs, DYNC1H1 is a large gene with 78 exons. Although our study revealed that DYNC1H1 variants were widely dispersed across the whole gene, it was notable that exons 4, 14, 5, and 8 were the mutational hot regions of DYNC1H1 after adjusting for exon length in this study. A previous study including 14 families with DYNC1H1 mutations found that 13 of the 14 families had mutations in the tail domain of DYNC1H1. Nevertheless, a cluster of mutations occurred in exon 8 within the tail domain itself (19). This conclusion might be due to the fact that exon 8 is the longest encoding exon of the DYNC1H1 gene, as our study also showed that exon 8 contained the most mutations while not adjusting for exon length. Despite the discrepancy between our study and previous research, all the mutational hot regions (including exons 4, 5, 8, and 14) were in the DHC/N1 region of the tail domain. The tail domain lies in the N terminal of the dynein heavy chain and contains binding positions for dynein intermediate chain (DIC) and dynein intermediate light chain (DILC), the latter of which further provides a separate binding site for dynein light chain (DLC). The DIC, DILC, and DLC together form the cargo-binding site of the dynein complex (20). Therefore, mutations in the tail domain affect the binding capacity of the dynein heavy chain with other components, which can lead to neurological impairment possibly via disrupting the retrograde transport of activated neurotrophin receptors (21, 22) and nerve injury signals.

**DISCUSSION**

**DYNC1H1** encodes heavy chain 1 of the dynein complex and is involved in the control of microtubule binding, as well as the recruitment of other dynein components (15, 18). In this study, we reported two SMALED1 cases with heterozygous DYNC1H1 mutations c.1792C>T (p.R598C) and c.790C>G (p.R264G). The diagnosis of SMALED1 was supported by neuroelectrophysiological examinations, which revealed chronic denervation and reinnervation manifestations with the relatively preserved function of nerve conduction. The relative lack in active denervation potential was in accordance with the chronic course of these patients. Through further genotype-phenotype analysis, we found that patients with mutations in the DYN1 region may have a more severe phenotype, more complicated clinical manifestations, and more CNS involvement than those who had mutations in the DHC/N1 region.

**TABLE 1** Comparison of mutation site distribution between SMALED1 and CMT2O families.

| Diagnosis | Total families | Mutation sites | Fisher exact test |
|-----------|----------------|----------------|------------------|
|           |                | DHC/N1 domain (%) | DYN1 domain (%) | P-value |
| SMALED1   | 43             | 35 (81.4)       | 8 (18.6)        | 0.488   |
| CMT2O     | 3              | 2 (66.7)        | 1 (33.3)        |         |

CMT2O, Charcot–Marie–Tooth type 2O; SMALED1, spinal muscular atrophy with lower extremity predominance 1.
FIGURE 3 | Genotype–phenotype correlation of DYNC1H1 mutations. (A) Correlations between mutations in different domains and clinical manifestations of neuromuscular diseases. The large red circle indicates that all patients carrying DYNC1H1 mutations in a specific domain have the relevant phenotype labeled on the left side. The small blue circle refers to a small subset of patients who have the phenotype. (B) Comparison of the distribution pattern of onset age between the (Continued)
(23, 24) or by an interruption in the clearance of misfolded proteins by autophagy (25). Of the mutational hot regions in this study, c.1792C>T was the most frequent point mutation, which was predicted to replace an arginine with a cysteine at the DIC-interacting site. However, no c.1792C>T mutation has been found in the East Asian population in previous publications. On the one hand, this discrepancy may reflect differences at the genetic level among different races; on the other hand, this result may be due to the relatively limited amount of cases in this area. Considering the large population basis in East Asia, more different kinds of DYNC1H1 mutations may be gradually detected, as we reported a teenage patient with SMALED1 caused by DYNC1H1 c.1792C>T heterozygous mutation in this study. Notably, c.751C>T (p.R251C) and c.2327C>T (p.P776L) were the most commonly reported mutations in East Asia. These two mutation sites lie in exon 4 and exon 8, which provides further support for the conclusion that these exons are hot regions of DYNC1H1 mutations. Recent years have witnessed significant improvement in gene sequencing techniques, and the potential of gene therapy has been trialed in treating various incurable hereditary diseases. Considering the highly dispersed distribution of DYNC1H1 mutations, precise base editing may not be practical for correcting a DYNC1H1 mutation in most patients; however, specific editing of the hotspots in the mutational hot regions has a promising future (26).

To our knowledge, few studies have been devoted to unraveling the genotype–phenotype relationship between neuromuscular disorders and DYNC1H1 variants. In the current study, mutations in the DYN1 region of DYNC1H1 protein were found to be correlated with more severe clinical phenotypes, more complicated manifestations, and more common CNS impairment than variants in the DHC_N1 region. This is in accordance with earlier findings that mutations in the tail domain (close to the N terminal) usually lead to pure motor neuron deficits hardly with any signs of brain abnormalities (4, 12), while mutations in the motor domain (close to the C terminal) often cause the malformation of cortical development (MCD) and epilepsy (15). The reasons underlying this phenotypic diversity have not been clearly elucidated yet. One study based on the yeast model system suggested that the degree of genetic dysfunction may be a potent determinant of phenotypic type. In this study, Matthew et al. revealed that motor system deficits seem to be susceptible to a small degree of dynein dysfunction, while brain function abnormalities appear to be associated with larger degrees of dysfunction (3). In precision, the coefficient of dynein dysfunction (CDD) for motor neuron diseases is from 5 to 18, smaller than the CDD for malformation of cortical development, which is ≥ 19 (3). Given that, in our study, different severity of clinical manifestations is correlated with different DYNC1H1 protein regions that mutations are located on, it is deduced that different mutational regions may lead to divergence in the degrees of a genetic dysfunction, which finally causes variability in the severity of dynein protein abnormalities. Further functional studies of the genetic characteristics at different sites of dynein protein will help us to explore the reason for the phenotypic heterogeneity.

The conclusions of this study should not be assessed without the consideration of several limitations. First, the study did not present a complete mutation spectrum since patients without any detailed description of clinical features were excluded from this study. Second, the reporting bias of included articles should not be ignored because cases with recurrent variants and classical phenotypes were less likely to be reported. Third, all these comparisons are based on the literature and online databases with a relatively limited amount of cases, and the genotype–phenotype correlations should be validated in larger patient population in future. Despite the limitations, our study provided a comprehensive summary of the DYNC1H1 mutation spectrum by selecting relevant neuromuscular diseases, especially SMALED1 and CMT2O, and unraveled the hidden correlation between genotypes and phenotypes.

CONCLUSION
In this study, we reported novel DYNC1H1 c.790C>G (p.R264G) heterozygous mutations that caused SMALED1. We provided a detailed description of the clinical and genetic spectrum of neuromuscular diseases caused by DYNC1H1 mutations by reviewing previous publications. Our findings also suggest that mutations in the DYN1 region of DYNC1H1 may be correlated with more severe clinical phenotypes, more complicated manifestations, and more common CNS involvement, which may shed light on unraveling the genetic mechanism underlying clinical heterogeneity of “dyneinopathy.”

DATA AVAILABILITY STATEMENT
The datasets presented in this study can be found in online repositories. The name of the repository and accession number(s) can be found below: National Center for Biotechnology Information (NCBI) GenBank, https://www.ncbi.nlm.nih.gov/genbank/, ON548480–ON548485.

ETHICS STATEMENT
The studies involving human participants were reviewed and approved by the Ethics Committee of Huashan Hospital, Fudan
University. Written informed consent was obtained from the individual(s), and minor(s)' legal guardian/next of kin, for the publication of any potentially identifiable images or data included in this article.

AUTHOR CONTRIBUTIONS

J-TL and S-QD completed the collection and assessment of previous publications independently and wrote the manuscript. X-JC revised the manuscript. All authors designed the study, collected the clinical and genetic data of the two patients, read, and approved the manuscript.

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SUPPLEMENTARY MATERIAL

The Supplementary Material for this article can be found online at: https://www.frontiersin.org/articles/10.3389/fneur.2022.943324/full#supplementary-material

Supplementary Figure 1 | Flow chart of publication screening process. A total of 22 previous publications out of 49 original articles were finally included according to the inclusion and exclusion criteria.

Supplementary Table 1 | DYNC1H1 variants associated with neuromuscular diseases in previous publications.

Supplementary Table 2 | Demographics and clinical characteristics of 105 patients with DYNC1H1 variants.

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