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JAMA Neurol. Author manuscript; available in PMC 2023 April 27.

Published in final edited form as:

JAMA Neurol. 2022 April 01; 79(4): 405–413. doi:10.1001/jamaneurol.2022.0067.

Multicenter Consensus Approach to Evaluation of Neonatal Hypotonia in the Genomic Era: A Review

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Author Contributions: Drs Morton and Agrawal had full access to all the data in the study and take responsibility for the integrity of the data and the accuracy of the data analysis.

Concept and design: Morton, Christodoulou, Muntoni, Raymond, Williams, Stark, Rowitch, Agrawal.

Acquisition, analysis, or interpretation of data: Morton, Costain, Wakeling, Wojcik, French, Szuto, Dowling, Cohn, Raymond, Darras, Lunke, Stark, Agrawal.

Drafting of the manuscript: Morton, Christodoulou, Muntoni, Wakeling, French, Cohn, Stark, Rowitch, Agrawal.

Critical revision of the manuscript for important intellectual content: Christodoulou, Costain, Wakeling, Wojcik, Szuto, Dowling, Cohn, Raymond, Darras, Williams, Lunke, Stark, Rowitch, Agrawal.

Statistical analysis: Morton.

Obtained funding: Raymond, Williams, Stark.

Administrative, technical, or material support: Morton, Wakeling, Cohn, Darras, Lunke, Agrawal.

Supervision: Muntoni, Cohn, Raymond, Darras, Stark, Rowitch, Agrawal.

Conflict of Interest Disclosures: Dr Christodoulou reported being a named investigator on a grant that involved next-generation sequencing of infants as part of an ultrarapid genomic testing research project; he reported receiving no direct funding through this project; however, some of the data generated in this project was included in the manuscript. Dr Muntoni reported receiving grants from Sarepta Therapeutics and Biogen and consultant fees from Sarepta Therapeutics, Pfizer, Novartis, Dyne, Biogen, and Roche outside the submitted work. Dr Wojcik reported receiving grants from the National Institutes of Health/National Institute of Child Health and Human Development during the conduct of the study. Dr Dowling reported receiving grants from Astellas for X-linked myotubular myopathy gene therapy research and medical advisory fees from Dynacure and Kate Therapuetics outside the submitted work; being the chair of the TREAT NMD executive committee; and being on the scientific advisory boards of the RYR1 Foundation, Muscular Dystrophy Association, and Muscular Dystrophy Canada. Dr Darras reported serving as an ad hoc scientific advisory board member for Audentes, AveXis/Novartis Gene Therapies, Biogen, Pfizer, Vertex and Roche/Genentech; serving as steering committee chair for the Roche FIREFISH study; being a data safety monitoring board member for Amicus Inc; receiving research support from the National Institutes of Health/National Institute of Neurological Disorders and Stroke, the Slaney Family Fund for spinal muscular atrophy, the Spinal Muscular Atrophy Foundation, CureSMA, and Working on Walking Fund; receiving research grants from Ionis Pharmaceuticals Inc, Biogen, Sarepta Pharmaceuticals, Novartis (AveXis), PTC Therapeutics, Roche, Scholar Rock, and Fibrogen; and receiving royalties for books and online publications from Elsevier and UpToDate Inc. Dr Rowitch reported having a contract with Illumina UK for whole-genome sequencing. Dr Agrawal reported being a member of the scientific advisory board of GeneDx and Illumina Inc. No other disclosures were reported.

Disclaimer: The views expressed are those of the authors and not necessarily those of the National Institute for Health Research or the UK Department of Health and Social Care.

Additional Contributions: We thank Antonio Garcia, PhD, University of Cambridge, Cambridge, United Kingdom, for figure preparation, and Tayyaba Khan, MPH, Sick Kids Research Institute, Toronto, Ontario, Canada, for assistance with data collection at the Toronto neonatal intensive care unit. No one was financially compensated for their contribution.

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Abstract

IMPORTANCE—Infants with hypotonia can present with a variety of potentially severe clinical signs and symptoms and often require invasive testing and multiple procedures. The wide range of clinical presentations and potential etiologies leaves diagnosis and prognosis uncertain, underscoring the need for rapid elucidation of the underlying genetic cause of disease.

OBSERVATIONS—The clinical application of exome sequencing or genome sequencing has dramatically improved the timely yield of diagnostic testing for neonatal hypotonia, with diagnostic rates of greater than 50% in academic neonatal intensive care units (NICUs) across Australia, Canada, the UK, and the US, which compose the International Precision Child Health Partnership (IPCHiP). A total of 74% (17 of 23) of patients had a change in clinical care in response to genetic diagnosis, including 2 patients who received targeted therapy. This narrative

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review discusses the common causes of neonatal hypotonia, the relative benefits and limitations of available testing modalities used in NICUs, and hypotonia management recommendations.

CONCLUSIONS AND RELEVANCE—This narrative review summarizes the causes of neonatal hypotonia and the benefits of prompt genetic diagnosis, including improved prognostication and identification of targeted treatments which can improve the short-term and long-term outcomes. Institutional resources can vary among different NICUs; as a result, consideration should be given to rule out a small number of relatively unique conditions for which rapid targeted genetic testing is available. Nevertheless, the consensus recommendation is to use rapid genome or exome sequencing as a first-line testing option for NICU patients with unexplained hypotonia. As part of the IPCHiP, this diagnostic experience will be collected in a central database with the goal of advancing knowledge of neonatal hypotonia and improving evidence-based practice.

The presentation of primary hypotonia in the neonatal intensive care unit (NICU) is complex to diagnose. Although our understanding of the genetic basis of hypotonia has advanced significantly in the past decade,[1–5] there remains a lag in implementation of state-of-the-art genetic testing along with variation in diagnostic approaches across institutions. Availability of effective treatments for genetic conditions, such as congenital myasthenic syndromes or spinal muscular atrophy (SMA),[6–8] highlights the importance of a timely diagnosis, and therapies for other hypotonic conditions will probably become available in the future. Therefore, prompt genetic diagnosis is increasingly informing clinical care decision and benefit from targeted treatments.[9–12]

Here, we have developed a consensus approach to genetic testing for infants with unexplained hypotonia. Experts volunteered from 5 medical centers that are members of the International Precision Child Health Partnership (IPCHiP): Royal Children's Hospital, Melbourne, Australia; The Hospital for Sick Children, Toronto, Ontario, Canada; Cambridge University Hospitals and Great Ormond Street Hospital, London, UK; and Boston Children's Hospital, Boston, Massachusetts. We systematically reviewed the diagnosis and outcomes of infants who presented with neonatal hypotonia during the period of 5 years (2016–2020) at each center. Based on iterative review of these data, we formulated an evaluative approach with consistent application of genomic testing in the NICU. In our experience, diagnostic rates by exome sequencing (ES) or genome sequencing (GS) for those infants exceeded 50%.

Clinicopathophysiological Observations

Pathophysiology

The causes of neonatal hypotonia are diverse, and the differential diagnosis is influenced by multiple factors, including pattern of hypotonia, family history, and accompanying signs and symptoms (multisystemic or hypotonia as the primary finding)[1] (eTable 1 in the Supplement). In contrast to weakness—defined as a reduction in maximum voluntary power of the muscles—hypotonia may be defined as reduced resistance to passive range of motion (phasic tone), or loss of postural control.[3] Here we will consider conditions characterized by hypotonia, which are often accompanied by weakness.

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Hypotonia can be primary or secondary. Primary causes of hypotonia can be categorized into broad categories of (1) central nervous system (CNS) disorders (central hypotonia; 60%–80% of primary neonatal hypotonia) and (2) peripheral nervous system (PNS) disorders, including neuromuscular disorders (15%–30% of primary neonatal hypotonia).[13] Secondary causes of hypotonia, such as hypoglycemia, congenital heart disease, and sepsis, are common and are routinely diagnosed during an initial evaluation. Therefore, when an obvious secondary cause is not identified, primary causes of hypotonia should be considered.

Clinical Presentation

CNS hypotonia manifests as low muscle tone, normal or exaggerated deep tendon reflexes, and relative preservation of antigravity movements. Primary genetic etiologies for CNS neonatal hypotonia include chromosomal abnormalities, such as trisomy 21 or Prader-Willi syndrome (PWS)[14,15]; neurometabolic conditions, such as peroxisomal disorders and other inborn errors of metabolism; malformations of cortical development; and a broad range of monogenic disorders where CNS dysfunction is considered a primary mechanism of disease (covered in recent reviews[2,3,16,17]). Non-genetic causes of CNS hypotonia include hypoxic-ischemic encephalopathy, infection, intracranial hemorrhage, CNS/spinal cord trauma, or craniocervical junction lesions, such as Chiari malformation. Encephalopathy is commonly observed in CNS-related causes of neonatal hypotonia.

PNS hypotonia is a major manifestation of neuromuscular disorders that can involve the anterior horn cells of the spinal cord (motor neurons), peripheral nerve, neuromuscular junction, or skeletal muscle. The PNS conditions, in contrast to CNS disorders, often (1) have hypotonia as the presenting symptom and (2) have absent or diminished deep tendon reflexes, absent neonatal reflexes, and profound weakness.

Conditions that reduce movements in utero can lead to joint contractures (arthrogryposis). Primary congenital muscle disorders include myopathies,[18] muscular dystrophies,[19] and myotonic dystrophies.[16,20] Congenital myopathies and muscular dystrophies typically present in the neonatal period with hypotonia, weakness, and arthrogryposis (Figure 1). Evidence of muscle injury, such as elevated creatine phosphokinase levels, is typical of congenital muscular dystrophy, and infants may have additional features, such as CNS, cardiac, or eye abnormalities. Congenital myotonic dystrophy type 1 (DM1) is often inherited from a mother with DM1 who has no or mild symptoms. Neonatal-onset SMA,[21] or SMA type 0 or 1A, is a rare but severe motor neuron disease which is rapidly progressive and is associated with proximal and axial weakness, as well as respiratory failure with typical paradoxical inspiratory movements owing to the relative sparing of the diaphragm. SMA 1B, the most common form, can also present within the first weeks of life. Other less common but treatable conditions are neuromuscular junction disorders, including aminoglycoside toxic effects, congenital myasthenic syndromes, and transient neonatal myasthenia from placental transfer of maternal antibodies against the acetylcholine receptor.

Congenital myasthenic syndromes are associated with primary dysfunction of the neuromuscular junction that manifest as fatigable weakness. Transient neonatal myasthenia presents with initial weakness with preservation of deep tendon reflexes. Peripheral

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neuropathies, characterized by distal weakness and atrophy, can have severe early-onset forms, such as congenital hypomyelinating neuropathy. PNS conditions that can be multisystemic include metabolic conditions, such as mitochondrial disorders, acid maltase deficiency, and fatty acid oxidation disorders. Such multisystemic disorders may be diagnosed through additional evaluation of metabolites such as lactic acid, plasma amino acids, plasma acylcarnitines, and urine organic acids.

Optimizing Genetic Diagnosis of Neonatal Hypotonia

Genetic causes of neonatal hypotonia include single-nucleotide variants (SNVs) or small insertion/deletion variants in the nuclear or mitochondrial genomes, expansions of repeated elements (as in congenital DM1), copy number variations, changes in DNA methylation states or uniparental disomy (as in PWS), and aneuploidies.[2,4,5,17,18,22–25] Clinically available genetic testing modalities each detect a different range of variants with variable reliability (Table 1; eTable 2 in the Supplement).[26–28] The optimal diagnostic approach for neonatal hypotonia, therefore, depends on the frequency of each genetic condition, as well as the relative costs and time to report of each test. Patients with neurodevelopmental disorders have higher diagnostic yields for GS than for microarray.[29,30] Further, mitochondrial genome sequencing, alone or as part of GS, may be needed to diagnose a disorder caused by a variant in the mitochondrial genome.[31–33]

Three common causes of neonatal hypotonia that have rapid targeted testing available should be considered early in evaluation: DM1, PWS, and SMA. DM1 is best detected by targeted testing; GS also offers the potential to detect repeated expansions. PWS can be caused by deletion of paternal genes, maternal uniparental disomy, or imprinting defects of genes in 15q11.2-q13[34]; PWS may be detected by chromosomal microarray (CMA), methylation testing, and/or detection of isodisomic regions via GS or ES.[14] Finally, diagnosis of SMA, caused by recessive variants of *SMN1* (most commonly deletion of exon 7), poses specific technical challenges, and reliable diagnosis requires targeted assays or optimization of genomic analysis pipelines. As early or presymptomatic treatment of SMA results in dramatically improved clinical outcomes compared with postsymptomatic onset of therapy, SMA testing is increasingly included in routine newborn screening.[35,36] For example, 35 states in the US have implemented newborn screening that can lead to the diagnosis of SMA during the first week of life and prompt treatment with the medications nusinersen or onasemnogene abeparvovec-xioi, both for SMA.

A multicenter database reflecting contemporary experience with neonatal hypotonia is needed to enhance collaboration and accelerate development of evidence-based guidelines. Toward this objective, a new collaborative to advance diagnosis and treatment for rare pediatric conditions called IPCHiP has combined the recent experiences in determining the genetic basis of hypotoniaamong5academic NICUs from 3 continents. Of the 5 hospitals, 4 are specialist referral centers, and 1 is a regional hospital. Royal Children’s Hospital has a 38-bed level 4 NICU and a 28-bed pediatric intensive care unit, admitting newborns with complex medical and surgical needs. Level 4 NICUs are also found at The Hospital for Sick Children (36 beds), Cambridge University Hospitals (58 beds), Great Ormond

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Street Hospital (9 beds), and Boston Children's Hospital (24 beds). The experiences of each institution contributing to this common analysis are detailed below:

- Royal Children's Hospital, Melbourne, Australia: The Royal Children's Hospital completed a pilot study led by the Victorian Clinical Genetics Services providing singleton rapid ES with a 2- to 3-week turnaround time (TAT) in 2016 to 2017.[37] The program then transitioned to ultrarapid trio ES (3-day TAT) in 2018 to 2019 and subsequently to ultrarapid trio GS (3-day TAT) in 2020.[38] All newborns presenting with isolated hypotonia underwent CMA, myotonic dystrophy testing, and SMA testing before ultrarapid genomic testing. The site is also a national referral center for ultrarapid genomic testing from throughout Australia as part of the Australian Genomics Acute Care Study.[38] Of the 108 patients published, 4 of 8 (50%) with hypotonia received a molecular diagnosis via ES.[38]
- The Hospital for Sick Children, Toronto, Ontario, Canada: At The Hospital for Sick Children, 44 infants had initially unexplained hypotonia from 2017 to 2020. In 2017, clinical genomic testing and extensive biochemical screening investigations were completed for 8 patients with hypotonia, of which 4 (50%) received a molecular diagnosis.[39]
- Cambridge University Hospitals, Cambridge, UK: Cambridge University Hospitals was the site of the Next Generation Children's Project, in which 159 patient-family trios received rapid TAT GS.[40] Of these, 3 of 10 patients (30%) with neonatal hypotonia in the cohort had pathogenic variants in*MTM1*(hemizygous), and a large de novo deletion in 5q15–23 (24 megabase) and SMA.
- Great Ormond Street Hospital, London, UK: At Great Ormond Street Hospital, rapid-trio ES is used for diagnosis of acutely unwell children, sometimes supplemented with targeted tested for PWS and SMA. No published results are available.
- Boston Children's Hospital, Boston, Massachusetts: At Boston Children's Hospital, rapid ES with TAT of 5 to7 business days[41] was completed for 15 patients with hypotonia. A diagnosis was reached with ES in 12 patients (80%). Ten of 15 infants (67%) had a single gene disorder. The 2 additional ES diagnoses were an infant with copy number variations (also detected by CMA) and an infant with PWS due to uniparental disomy. One infant with negative findings on ES needed mitochondrial GS to diagnose Leigh syndrome. In addition, 2 infants had a variant of unknown significance (VUS) that needed further workup to attribute pathogenicity.

Each IPCHiP center hospital reviewed all patients cared for during their recent experiences from prior publications who had undergone an ES/GS evaluation for primary hypotonia after exclusion of conditions such as trisomy 21. All hospitals have implemented genomic testing early in the care of patients with hypotonia. Our results indicate that 56% of neonates (23 of 41) with hypotonia received a genetic diagnosis by ES/GS (Table 2). When separately considering the 12 infants who have primary hypotonia compared with those with

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multisystemic findings, such as seizures or structural brain abnormalities, 10 (83%) had a genetic diagnosis, which included 4 (40%) with congenital myopathy, 4 (40%) with PNS disorder (including 3 with SMA), 1 (10%) with a syndrome that included myopathy, and 1 (10%) with PWS (eTables 3 and 4 in the Supplement).Our experience indicates that the diagnosis can be significantly expedited through use of trio ES/GS, where both parents' sequences can be compared with the infant's sequence. Trio sequencing allows rapid and more efficient prioritization of variants of interest and can provide more definitive variant classifications by establishing inheritance patterns, thereby reducing uncertainty and time to diagnosis.

Treatment

Infants affected by hypotonia can require prolonged life-supporting care, such as assisted ventilation and nutritional support. Further, many infants require invasive testing, such as skin or muscle biopsy or electromyography, lumbar puncture, or invasive medical procedures (eg, tracheostomy and gastrostomy placement). As testing times continue to improve, molecular diagnoses can shorten the length of hospital stay for patients with hypotonia and inform management choices. In our case series, 74% of patients (17 of 23) for whom detailed clinical information was available had a change in clinical care after genetic diagnosis, and 2 received targeted therapy for mitochondrial DNA depletion syndrome and congenital myasthenic syndrome. Of the 5 patients who did not have a change in clinical care, 1 (20%) had results return after death, 2 (40%) were referred to research opportunities, and in 2 families, the diagnosis informed reproductive options for future pregnancies. Gene therapy is being developed to treat several causes of neonatal hypotonia, including SMA, *MTM1*-related congenital myopathy, aromatic L-amino acid decarboxylase deficiency, giant axonal neuropathy, and metachromatic leukodystrophy.[10–12,42]Molecular-based precision therapies also include antisense oligonucleotides, currently in clinical practice for SMA, tested in other conditions in an n-of-1 setting[43] and in consideration for a range of diseases including infantile epileptic encephalopathies. For several infants, early initiation of precise therapy for these disorders appears to provide added benefit, making timely diagnosis essential to improve patient outcomes. Additional disease-modifying therapies, such as enzyme replacement therapy, are also used either in clinical practice (eg, for Pompe disease) or are under development (eg, for forms of neuronal ceroid lipofuscinosis). Furthermore, early diagnosis of a few genetic metabolic disorders, with prompt implementation of specific therapy, can greatly ameliorate the infant's clinical condition. Prognostic information, such as the anticipated trajectory of severe conditions without available treatments, is important to share with families that may help with potential palliative care decisions.

Discussion

Efficient diagnosis of rare mendelian disorders is essential to advancing treatments.[44] We developed IPCHiP, an international consortium of academic centers across Australia, Canada, UK, and US, dedicated to promoting the role of genomic medicine in pediatrics. As part of this collaboration, we are developing an international patient database to enable evidence-based diagnostic recommendations, consistent with our overarching goal of bringing innovative solutions to the management of patients with rare diseases. Our

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proposed recommendations for evaluation of hypotonia (Figure 2) are based on the experience and expertise of member sites of IPCHiP and emphasize the role of ES or GS early in the diagnostic pathway with the view that such information will optimize patient management. In our experience, use of ES/GS early in the evaluation process improves diagnostic rates and leads to meaningful changes in care. Although in this review we only describe case summaries that were previously published of patients with hypotonia who had ES/GS, more recent clinical experience at our institutions with larger numbers of patients suggests a similar diagnostic yield of 50% or greater.

Microarray analysis and/or targeted testing for trisomy 21, PWS, SMA, and DM1 are important to consider during the initial evaluation as those results can be returned rapidly and influence care decisions. Clinical phenotyping, driven by the patient's features, is essential to aid interpretation of the genetic test. Although many NICUs use multigene panels to evaluate neonatal hypotonia or use ES/GS after a substantial number of other investigations have been completed, our experience argues that rapid-diagnostic trio ES/GS analysis is the preferred first-line diagnostic test. Rapid testing provides the greatest benefit, both for mitigating prolonged and invasive diagnostic testing (eg, electromyography, muscle biopsy), optimizing medical management, and decreasing intensive care costs.[29,45,46] Although advancements in bioinformatic analysis of ES/GS have shown increased potential for detecting structural variants (eg, SMA), repeated expansions (eg, DM1), and uniparental disomy (eg, PWS), criterion-standard targeted testing should be considered if the clinical suspicion of these disorders is high. This decision will also be influenced by the scope of local ES/GS diagnostic pipeline validation, relative costs, and TAT. Given the greater diagnostic potential of GS, in the future, IPCHiP will focus on enhancing the evidence base and clinical implementation of GS.

There is a high prevalence of VUS and a concomitant substantial unmet need in diagnostic testing.[47–49] VUS can be characterized with predictive algorithms that assess interspecies conservation, protein domain structure,[50] and variation within reference human cohorts[51] to infer functional consequences. Guidelines that discuss the factors relevant to interpretation of VUS in clinical scenarios have been developed.[49,52] The effect of VUS may be unclear either owing to lack of information about the role of genes in human disease, or the unknown effect of specific variants on disease gene function.[53] Coordinating an expert multidisciplinary review of VUS can increase sharing of knowledge resources across different health care systems and facilitate patient-oriented research that moves variants into more definitive functional categories, thereby increasing diagnostic yield. As knowledge advances, periodic reevaluation of VUS is indicated.

Our group comprises experts in neuromuscular, motor neuron, and glial pathobiology to support extending our scope from the bedside to the laboratory. Functional modeling of VUS[54,55] and validation of the pathological consequences of these DNA changes, such as assessing the effect on production of relevant protein(s) or RNA splicing in muscle or skin biopsies, or the introduction of VUS in patient-derived–induced pluripotent or mesenchymal stem cells,[56–59] could provide functional data to support a genetic diagnosis. Additionally, such models could be used to develop novel therapeutics. Important technical issues remain unaddressed in the area of VUS interrogation, including limited ability to

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predict the functional effect of missense and noncoding variants, lack of resources for cell-specific functional validation, and lack of rapid model organism approaches to assay the effect of VUS during development. In addition, our consortium will incorporate and evaluate emerging diagnostic modalities, such as RNA sequencing, proteomics, and DNA methylomics. Early data suggest that RNA sequencing can significantly increase diagnostic yield in pediatric monogenic disorders and can enable clarification of some VUS.[60,61] The potential utility of approaches, such as proteomics, require further study, as was demonstrated in a family with congenital myopathy and *PLIN4* repeated expansion.[62] By developing a core of experts with relevant domain knowledge and technical expertise, new collaborations can be forged to address these open questions.

Emerging therapies for neuromuscular disorders include antisense oligonucleotides, which bind to pre–messenger RNA and alter splicing to increase, restore, or reduce gene expression,[43] gene replacement via integrating or nonintegrating viral vector platforms, and clustered regularly interspaced short palindromic repeats–based technologies to edit the genome.[63] One of our main goals is to identify patients who can benefit from early personalized therapies. For SMA, there are gene replacement (onasemnogene abeparvovec-xioi),[10,64–67] antisense oligonucleotides (nusinersen),[7,8,68] and small molecule therapies.[69] The 2 clinical trials for congenital myopathy listed at ClinicalTrials.gov are gene transfer via adeno-associated virus for X-linked *MTM1* myotubular myopathy (AT132)[70] and antisense oligonucleotide knockdown of *DNM2* for centronuclear myopathy (DYN101),[71] although neither is currently enrolling neonates (Table 3). Other neuromuscular conditions that may be amenable to personalized therapies in the near future include specific pharmacological therapies for rare mitochondrial disorders[72] and gene replacement in rare infantile cardiomyopathies owing to *MYBPC3* variants.[73] To accelerate the development and implementation of new interventions, we will continue to evaluate patients for new therapeutic options.

Genome-wide testing (ES/GS) of infants can raise concerns about privacy, autonomy, and potential for misuse or discrimination.[74] These general concerns need to be considered appropriately and balanced against the potential direct benefits to patients of achieving a diagnosis in clinical presentations with high a priori risk. Parental acceptance of rapid genome-wide testing for intensive care unit populations is generally high[75] with enrollment rates of 80% or more at several institutions.[38,40,41] Genomic testing is increasingly recognized as a critical diagnostic test by health care professionals and families.[74] Nevertheless, rapid genomic testing in the already stressful environment of the NICU raises significant ethical and counseling issues[76–79] with long-term psychosocial effects on families, an important area for future research.

Conclusions

The diagnosis and care of neonates with hypotonia benefits from prompt and comprehensive genetic testing. Based on the experience of the IPCHiP consortium, in this narrative review, we propose a shift toward the use of rapid genomic testing as a first-tier test in this patient group with the aim of optimizing medical management and access to emerging therapies.

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Supplementary Material

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Funding/Support:

This work was supported in part by grant R01 AR068429 from the National Institute of Arthritis and Musculoskeletal and Skeletal Diseases of the National Institutes of Health and grant U19 HD077671 from the National Institute of Child Health and Human Development/National Human Genome Research Institute of the National Institutes of Health (Dr Agrawal); the Gene Discovery Core of The Manton Center for Orphan Disease Research, Boston Children's Hospital; the Boston Children's Hospital Office of Faculty Development Career Development Fellowship, American Heart Association Postdoctoral Fellowship, and American Heart Association Career Development Award (Dr Morton); grant BRC-1215-20014 from the National Institute for Health Research Cambridge Biomedical Research Centre and the National Institute for Health Research Rare Disease Bioresource; grant 1113531 from the National Health and Medical Research Council (Drs Stark, Lunke, and Christodoulou); the Royal Children's Hospital Foundation; grant GHFM76747 from the Medical Research Futures Fund; the Victorian Government's Operational Infrastructure Support Program; the Royal Children's Hospital Foundation (Dr Christodoulou); the Sick Kids Centre for Genetic Medicine; the University of Toronto McLaughlin Centre; and the National Institute for Health Research Great Ormond Street Hospital Biomedical Research Centre, the Great Ormond Street Institute of Child Health, the University College London, and the Great Ormond Street Hospital Trust (Dr Muntoni).

Role of the Funder/Sponsor:

The funders had no role in the design and conduct of the study; collection, management, analysis, and interpretation of the data; preparation, review, or approval of the manuscript; and decision to submit the manuscript for publication.

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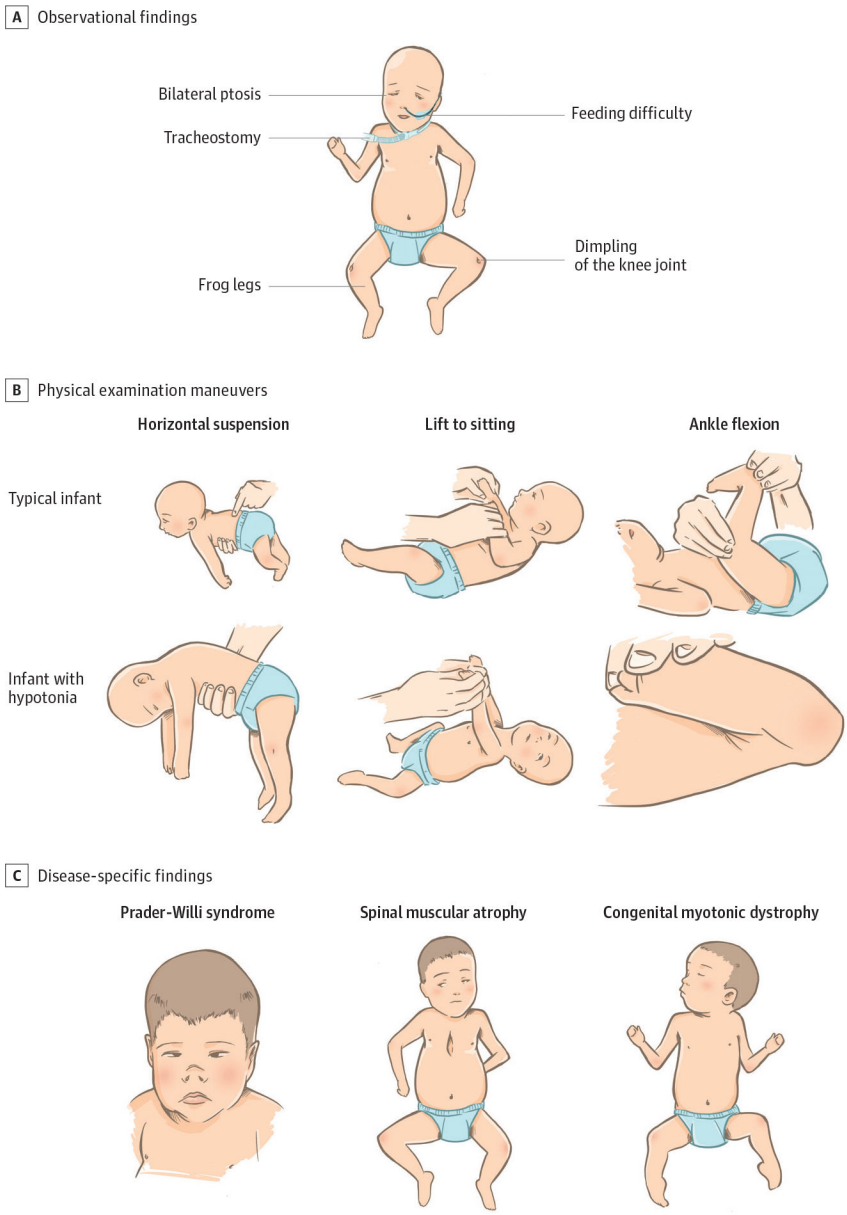

Figure 1. Physical Examination Findings in Neonatal Hypotonia

A, Observational findings during examination of infants with primary hypotonia can include frog-leg positioning owing to low tone and paucity of movement, the presence of a nasogastric feeding tube owing to poor feeding, tracheostomy for respiratory failure, knee dimpling from restricted movements in utero, and ptosis with myopathic facies in neuromuscular junction disorders. B, Physical examination maneuvers including horizontal suspension, lift to sitting position, and ankle flexion demonstrate differences between typical infants and infants with hypotonia. C, Prader-Willi syndrome can be accompanied by narrow nasal bridge, narrowing of the forehead at the temples, almond-shaped eyes, thin upper lip, and down-turned corners of the mouth. Infants with spinal muscular atrophy can demonstrate bell-shaped chest, retractions with respirations, paucity of movement, and

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passive positioning with flexed hips and knees. Congenital muscular dystrophy is often associated with joint contractures.

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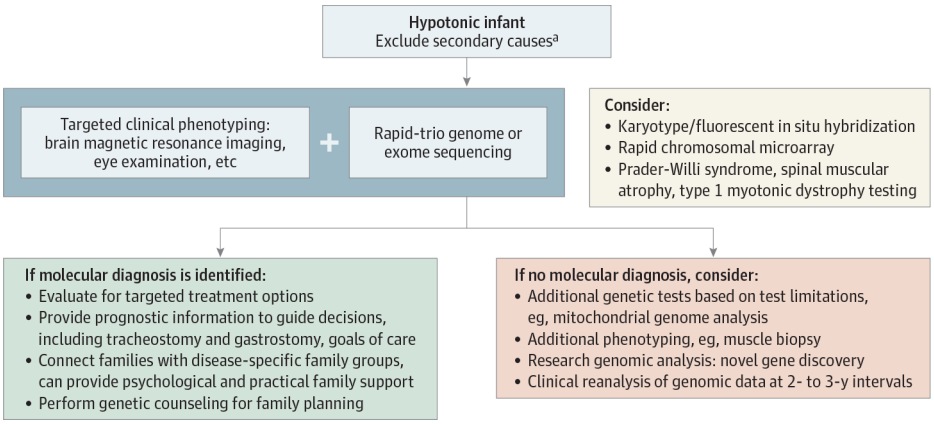

Figure 2. Consensus Diagnostic Algorithm for Neonatal Hypotonia

Evaluation of infants with hypotonia should include early exome of genome sequencing. Clinicians should also consider concomitant chromosomal microarray if other anomalies present, spinal muscular atrophy if not included in newborn screening, or type 1 myotonic dystrophy if evidence of myotonia in the mother.

[a] Ensure evaluation for secondary causes, such as infectious disease, hypoglycemia, inborn errors of metabolism, and hypoxic ischemic encephalopathy.

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Table 1.

Broad Categories of Genetic Testing Modalities for Hypotonia

Test	Aneuploidy	Large intergenic deletions/ duplications	Intragenic deletions/ duplications	Monogenic SNVs and small I/Ds	Repeated element expansions[a]	Methylation changes
Karyotype or fluorescence in situ hybridization (chromosome number and identity)	Optimaltest	Variable detection	Unable to detect	Unable to detect	Unable to detect	Unable to detect
Chromosomal microarray	Able to detect	Optimal test	Limited detection	Unable to detect	Unable to detect	May detect uniparental disomy or deletion leading to imprinting disorder
Next-generation sequencing-based gene panel	Unable to detect	Variable detection	Able to detect	Optimaltest	Unable to detect	Unable to detect
Methylation array or bisulfite sequencing (methylation state of DNA)	Unable to detect	Unable to detect	Unable to detect	Unable to detect	Unable to detect	Optimal test
Mitochondrial sequencing (mitochondrial genome sequencing)	Unable to detect	Unable to detect	Unable to detect	Optimaltest for mitochondrial genome SNV or I/Ds	Unable to detect	Unable to detect
Exome sequencing (autosomal coding sequences)	Able to detect	Able to detect	Able to detect	Optimal test	Limited detection	May detect uniparental disomy or deletion leading to imprinting disorder
Genome sequencing (coding and noncoding sequences)	Able to detect	Optimal test	Optimal test	Optimal test	Able to detect	May detect uniparental disomy or deletion leading to imprinting disorder

Abbreviations: I/D, insertion/deletion; SNV, single-nucleotide variant.

[a]Does not include targeted testing.

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Table 2.

Neonatal Intensive Care Unit Congenital Hypotonia Diagnosis by Exome Sequencing (ES) and Genome Sequencing (GS)

Center	Test	Patients with hypotonia, No.	Genetic diagnoses, No. (%)	Source
Boston Children's Hospital, Boston, Massachusetts	Trio ES	15	12 (80)	Gubbels et al,[41] 2020
Cambridge Children's Hospital, Cambridge, UK	Trio GS	10	3(30)	French et al,[40] 2019
Royal Children's Hospital, Melbourne, Australia	Trio ES/GS	8	4(50)	Stark et al,[37] 2018; Lunke et al,[38] 2020
The Hospital for Sick Children, Toronto, Ontario, Canada	ES/GS	8	4(50)	Djordjevic et al,[39] 2020
Total	NA	41	23 (56)	NA

Abbreviation: NA, not applicable.

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Table 3.

Examples of Emerging Therapies for Hypotonia

Therapy	Exampledrug (disease)	Mechanism	ClinicalTrials.gov identifier
Existing therapy			
Antisense oligonucleotides	Nusinersen (spinalmuscular atrophy)	Alter *SMN2* pre-mRNA splicing to produce increased full-length protein	NCT01839656[68]
Adeno-associated virus	Onasemnogene abeparvovec-xioi (spinal muscular atrophy)	Adeno-associated virus 9 delivery offunctional *SMN1* gene	NCT03306277[67]
Clinical trials			
Antisense oligonucleotides	DYN101 (centronuclear myopathy)	Inhibits *DNM2* expression by binding to pre-mRNA	NCT04033159[71]
Adeno-associated virus	AT132 (X-linked myotubular myopathy)	Adeno-associated virus 8 delivery offunctional *MTM1* gene	NCT03199469[70]

Abbreviation: mRNA, messenger RNA.