Australian Government

Department of Health

Application Form

Genomic testing for the diagnosis of neuromuscular disorders

This application form is to be completed for new and amended requests for public funding (including but not limited to the Medicare Benefits Schedule (MBS)). It describes the detailed information that the Australian Government Department of Health requires in order to determine whether a proposed medical service is suitable.

Please use this template, along with the associated Application Form Guidelines to prepare your application. Please complete all questions that are applicable to the proposed service, providing relevant information only. Applications not completed in full will not be accepted.

Should you require any further assistance, departmental staff are available through the Health Technology Assessment Team (HTA Team) on the contact numbers and email below to discuss the application form, or any other component of the Medical Services Advisory Committee process.

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Email: hta@health.gov.au
Website: www.msac.gov.au

PART 1 – APPLICANT DETAILS

1. Applicant details (primary and alternative contacts)

Corporation / partnership details (where relevant):
Corporation name: The Royal College of Pathologists of Australasia
ABN: 52 000 173 231
Business trading name: The Royal College of Pathologists of Australasia

Primary contact name: REDACTED

Primary contact numbers
Business: **REDACTED**
Mobile: **REDACTED**
Email: **REDACTED**

Alternative contact name: REDACTED

Alternative contact numbers
Business: **REDACTED**
Mobile: **REDACTED**
Email: **REDACTED**

Alternative contact name: REDACTED

Alternative contact numbers
Business:
Mobile: **REDACTED**
Email: **REDACTED**

2. (a) Are you a lobbyist acting on behalf of an Applicant?

☐ Yes
☒ No

(b) If yes, are you listed on the Register of Lobbyists?

☐ Yes
☐ No

PART 2 – INFORMATION ABOUT THE PROPOSED MEDICAL SERVICE

3. Application title

Genetic testing for the diagnosis of neuromuscular disorders.

4. Provide a succinct description of the medical condition relevant to the proposed service (no more than 150 words – further information will be requested at Part F of the Application Form)

Neuromuscular disorders (NMDs) are a broad range of disorders. NMDs can be roughly allocated into four categories: muscle disorders such as Duchenne muscular dystrophy (DMD); motor neuron disorders including spinal muscular atrophies (SMAs); neuropathies such as Charcot-Marie-Tooth disease (CMT); and neuromuscular junction disorders (Arnold & Flanigan 2012). They have a high level of clinical and genetic heterogeneity, and overlapping phenotypes (Fattahi et al 2017). Many NMDs present antenatally or in early infancy and are associated with significant disability or life-threatening complications. NMDs tend to be genetic in origin, can be inherited as autosomal dominant, autosomal recessive, X-linked, or mitochondrial traits, however, *de novo* pathogenic variants are also common (Laing 2012). While historically treatment options for NMD were poor, new developments offer curative interventions or decrease morbidity and mortality (Dowling et al 2018). Many of the new treatments for NMDs are guided by establishment of a definitive genetic diagnosis.

5. Provide a succinct description of the proposed medical service (no more than 150 words – further information will be requested at Part 6 of the Application Form)

i. **Gene panel testing to identify pathogenic variants for genetic neuromuscular disorders in patients where clinical criteria or a family history indicate that genetic testing is warranted:** As NMDs can be difficult to categorise, broad panels of genes sequenced by next generation sequencing (NGS) are considered the best option in order to capture as many potential causative variants as possible. *Fewer genes on a panel would result in a reduced diagnostic rate.* Based on clinical criteria, patients will be triaged to undergo testing with *either* a myopathy or neuropathy panel.

ii. **Cascade testing of family members:** Mutation-specific detection of a clinically actionable pathogenic variant previously identified in a biological relative.

iii. **Prenatal genetic testing** using mutation-specific detection of a clinically actionable pathogenic variant(s) previously identified in a relative.

iv. **Prenatal genetic testing** for NMD using gene panels: for a suspected NMD in the prenatal period with no previous genetic testing performed in the family, after appropriate counselling.

6. (a) Is this a request for MBS funding?

☒ Yes
☐ No

(b) If yes, is the medical service(s) proposed to be covered under an existing MBS item number(s) or is a new MBS item(s) being sought altogether?

☐ Amendment to existing MBS item(s)
☒ New MBS item(s)

(c) If an amendment to an existing item(s) is being sought, please list the relevant MBS item number(s) that are to be amended to include the proposed medical service:

N/A

(d) If an amendment to an existing item(s) is being sought, what is the nature of the amendment(s)?

i. ☐ An amendment to the way the service is clinically delivered under the existing item(s)
ii. ☐ An amendment to the patient population under the existing item(s)
iii. ☐ An amendment to the schedule fee of the existing item(s)
iv. ☐ An amendment to the time and complexity of an existing item(s)
v. ☐ Access to an existing item(s) by a different health practitioner group

vi. ☐ Minor amendments to the item descriptor that does not affect how the service is delivered
vii. ☐ An amendment to an existing specific single consultation item
viii. ☐ An amendment to an existing global consultation item(s)
ix. ☐ Other (please describe below):

(e) If a new item(s) is being requested, what is the nature of the change to the MBS being sought?

i. ☐ A new item which also seeks to allow access to the MBS for a specific health practitioner group
ii. ☒ A new item that is proposing a way of clinically delivering a service that is new to the MBS (in terms of new technology and / or population)
iii. ☐ A new item for a specific single consultation item
iv. ☐ A new item for a global consultation item(s)

(f) Is the proposed service seeking public funding other than the MBS?

☐ Yes
☒ No

(g) If yes, please advise:

N/A

7. What is the type of service:

☐ Therapeutic medical service
☒ Investigative medical service
☐ Single consultation medical service
☐ Global consultation medical service
☐ Allied health service
☐ Co-dependent technology
☐ Hybrid health technology

8. For investigative services, advise the specific purpose of performing the service *(which could be one or more of the following)*:

i. ☐ To be used as a screening tool in asymptomatic populations
ii. ☒ Assists in establishing a diagnosis in symptomatic patients
iii. ☒ Provides information about prognosis
iv. ☒ Identifies a patient as suitable for therapy by predicting a variation in the effect of the therapy
v. ☐ Monitors a patient over time to assess treatment response and guide subsequent treatment decisions
vi. ☒ A service that tests for heritable mutations in clinically affected individuals to make a genetic diagnosis and thus estimate their variation in (predisposition for) future risk of further disease and, when also appropriate, cascade testing of family members of those individuals who test positive for one or more relevant mutations, to make a genetic diagnosis and thus estimate each family member's variation in (predisposition for) future risk of developing the clinical disease

9. Does your service rely on another medical product to achieve or to enhance its intended effect?

☐ Pharmaceutical / Biological
☐ Prosthesis or device
☒ No

10. (a) If the proposed service has a pharmaceutical component to it, is it already covered under an existing Pharmaceutical Benefits Scheme (PBS) listing?

☐ Yes
☐ No

(b) If yes, please list the relevant PBS item code(s):

N/A

(c) If no, is an application (submission) in the process of being considered by the Pharmaceutical Benefits Advisory Committee (PBAC)?

☐ Yes (please provide PBAC submission item number below)
☐ No

N/A

(d) If you are seeking both MBS and PBS listing, what is the trade name and generic name of the pharmaceutical?

Trade name:
Generic name:

11. (a) If the proposed service is dependent on the use of a prosthesis, is it already included on the Prostheses List?

☐ Yes
☐ No
N/A

(b) If yes, please provide the following information (where relevant):

Billing code(s):
Trade name of prostheses:
Clinical name of prostheses:
Other device components delivered as part of the service:

(c) If no, is an application in the process of being considered by a Clinical Advisory Group or the Prostheses List Advisory Committee (PLAC)?

☐ Yes
☐ No

(d) Are there any other sponsor(s) and / or manufacturer(s) that have a similar prosthesis or device component in the Australian market place which this application is relevant to?

☐ Yes
☐ No

(e) If yes, please provide the name(s) of the sponsor(s) and / or manufacturer(s):

N/A

12. Please identify any single and / or multi-use consumables delivered as part of the service?

Single use consumables: General single use laboratory consumables such as pipette tips, centrifuge tubes etc
Multi-use consumables: Nil

PART 3 – INFORMATION ABOUT REGULATORY REQUIREMENTS

The National Association of Testing Authorities (NATA) and the Royal College of Pathologists Australasia (RCPA) oversee the regulation of massively parallel sequencing for clinical purposes. Laboratories require accreditation by a joint NATA/RCPA process to ISO 15189, and specifically accredited to provide genetic testing via massively parallel sequencing. This accreditation process covers the technical aspects of the laboratory sequencing, analysis pipelines, curation (or interpretation) of results and production of the report to a clinical standard. This allows any accredited laboratory to provide equivalent variant analysis services to a minimum standard. There are no requirements for use of specific manufacturer's reagents, equipment or analysis pipelines.

Note: A non-commercial IVD is required to be regulated but not to be listed on the ARTG: testing using an IVD would be delivered only by Approved Practising Pathologists in NATA Accredited Pathology Laboratories (as defined in MBS Pathology table) by referral only by registered Medical Practitioners (non-pathologists) in line with other tests in the MBS Pathology Table.

13. (a) If the proposed medical service involves the use of a medical device, in-vitro diagnostic test, pharmaceutical product, radioactive tracer or any other type of therapeutic good, please provide the following details:

Type of therapeutic good: In-vitro diagnostic test
Manufacturer's name: N/A
Sponsor's name: N/A

(b) Is the medical device classified by the TGA as either a Class III or Active Implantable Medical Device (AIMD) against the TGA regulatory scheme for devices?

☒ Class III
☐ AIMD
☐ N/A

14. (a) Is the therapeutic good to be used in the service exempt from the regulatory requirements of the *Therapeutic Goods Act 1989*?

☐ Yes (If yes, please provide supporting documentation as an attachment to this application form)
☒ No

(b) If no, has it been listed or registered or included in the Australian Register of Therapeutic Goods (ARTG) by the Therapeutic Goods Administration (TGA)?

☐ Yes (if yes, please provide details below)
☒ No

ARTG listing, registration or inclusion number:
TGA approved indication(s), if applicable:
TGA approved purpose(s), if applicable:

15. If the therapeutic good has not been listed, registered or included in the ARTG, is the therapeutic good in the process of being considered for inclusion by the TGA?

☐ Yes (please provide details below)
☒ No

Date of submission to TGA:
Estimated date by which TGA approval can be expected:
TGA Application ID:
TGA approved indication(s), if applicable:
TGA approved purpose(s), if applicable:

16. If the therapeutic good is not in the process of being considered for listing, registration or inclusion by the TGA, is an application to the TGA being prepared?

☐ Yes (please provide details below)
☒ No

Estimated date of submission to TGA:
Proposed indication(s), if applicable:
Proposed purpose(s), if applicable:

PART 4 – SUMMARY OF EVIDENCE

17. **Provide an overview of all key journal articles or research published in the public domain related to the proposed service that is for your application (limiting these to the English language only).** ***Please do not attach full text articles, this is just intended to be a summary.***

	Type of study design*	**Title of journal article or research project (including any trial identifier or study lead if relevant)**	**Short description of research (max 50 words)****	**Website link to journal article or research (if available)**	**Date of publication*****
1.	Cost-effectiveness analysis Australia	Cost-effectiveness of massively parallel sequencing for diagnosis of paediatric muscle diseases(Schofield et al 2017)	In 56 undiagnosed patients with childhood-onset muscle disorders, both NMD targeted gene panel and WES had increased diagnostic yields (from 46 to 75% for NMD panel, and 79% for WES), compared to muscle biopsy. The cost per diagnosis was reduced from US$16,495 to US$3,706 for the NMD panel and US$5,646 for WES.	https://www.nature.com/articles/s41525-017-0006-7.pdf	2017
2.	Cohort Australia	Next generation sequencing in a large cohort of patients presenting with neuromuscular disease before or at birth (Todd et al 2015)	Contemporary multi-gene sequencing (NMD gene panel or WES) was used for 45 patients with fetal akinesia/hypokinesia, arthrogryposis or severe congenital myopathies from 38 unrelated Australian families A conclusive genetic diagnosis was achieved for 18 of the 38 families (47%).	https://www.ncbi.nlm.nih.gov/pmc/articles/PMC4650299/pdf/13023_2015_Article_364.pdf	2015
3.	Diagnostic yield USA	A Comprehensive Genomic Approach for Neuromuscular Diseases Gives a High Diagnostic Yield (Ankala et al 2015)	Comparative studies for various genetic testing approaches indicated that NMD comprehensive panel testing has a 3-fold greater diagnostic yield (46%) than single gene testing (15–19%) and some variation may be missed by whole exome sequencing (about 18%). Targeted NMD panel is recommended as a first-tier test due to the highest clinical diagnostic yield for NMD.	https://onlinelibrary.wiley.com/doi/pdf/10.1002/ana.24303	2015

	Type of study design*	Title of journal article or research project (including any trial identifier or study lead if relevant)	Short description of research (max 50 words)**	Website link to journal article or research (if available)	Date of publication***
4.	Diagnostic yield Iran	Improved diagnostic yield of neuromuscular disorders applying clinical exome sequencing in patients arising from a consanguineous population (Fattahi et al 2017)	45 patients, mostly offspring of consanguineous marriages were examined using whole exome sequencing. Data analysis was performed to identify the most probable pathogenic rare variants in known NMD genes which led to the identification of causal variants for 33 out of 45 patients (73.3%).	https://onlinelibrary.wiley.com/doi/pdf/10.1111/cge.12810	2017
5.	Diagnostic yield USA	Diagnostic utility of exome sequencing in the evaluation of neuromuscular disorders (Haskell et al 2018)	Exome sequencing was performed in 93 patients with adult-onset NMDs. The overall incremental diagnostic yield of exome sequencing in the cohort was 12.9%, likely reflecting the less clear underlying aetiology in the later onset NMD.	https://www.ncbi.nlm.nih.gov/pmc/articles/PMC5798313/pdf/NG2017006239.pdf	2018
6.	Diagnostic yield Japan	Target resequencing of neuromuscular disease-related genes using next-generation sequencing for patients with undiagnosed early-onset neuromuscular disorders (Kitamura et al 2016)	42 Japanese patients referred with symptoms of muscle weakness and hypotonia neonatally and/or during childhood, and neuromuscular disorders. Muscle biopsy was performed in 16 of the 42 patients. For 34 patients, a definitive diagnosis was not yielded by conventional genetic tests. Causative genes were identified in 19/42 (45.2%) patients.	https://www.nature.com/articles/jhg201679.pdf	2016
7.	Diagnostic yield Taiwan	Expanding genotype/phenotype of neuromuscular diseases by comprehensive target capture/NGS (Tian et al 2015)	Thirty-five unrelated NMD families (38 patients) with clinical and/or muscle pathologic diagnoses but without identified causative genetic defects were analysed. Deleterious mutations were found in 29 families (83%). Definitive causative mutations were identified in 21 families (60%) and likely diagnoses were established in 8 families (23%).	https://www.ncbi.nlm.nih.gov/pmc/articles/PMC4807910/pdf/NG2015000224.pdf	2015

	Type of study design*	Title of journal article or research project (including any trial identifier or study lead if relevant)	Short description of research (max 50 words)**	Website link to journal article or research (if available)	Date of publication***
8.	Diagnostic yield Germany	Early-Onset Myopathies: Clinical Findings, Prevalence of Subgroups and Diagnostic Approach in a Single Neuromuscular Referral Center in Germany (Vill et al 2017)	98 patients with clinical symptoms suggestive of NMD underwent genetic testing. The final diagnosis could be found in 63 out of 98 patients (64%) with molecular genetic analysis. In 55% targeted gene sequencing could establish the genetic diagnosis.	https://content.iospress.com:443/download/journal-of-neuromuscular-diseases/jnd170231?id=journal-of-neuromuscular-diseases%2Fjnd170231	2017
9	Diagnostic yield Korea	Utility of next generation sequencing in genetic diagnosis of early onset neuromuscular disorders (Chae et al 2015)	43 patients presenting with early onset neuromuscular disorders from unknown genetic origin were tested by NGS for 579 nuclear genes associated with myopathy. In 21 of the 43 patients, the definite genetic causes were identified (48.8%).	https://jmg.bmj.com/content/jmedgenet/52/3/208.full.pdf	2015
10	Diagnostic yield	Next-generation sequencing-based molecular diagnosis of neonatal hypotonia in Chinese Population (Wang et al 2016)	In 186 neonates with hypotonia, the authors identified the genetic causes for 117 neonates by traditional detection methods or targeted NGS, achieving a high solving rate of 62.9%.	https://www.nature.com/articles/srep29088	2016

NMD = neuromuscular disease, MPS = massive parallel sequencing, WES = whole exome sequencing

** Categorise study design, for example meta-analysis, randomised trials, non-randomised trial or observational study, study of diagnostic accuracy, etc.*

***Provide high level information including population numbers and whether patients are being recruited or in post-recruitment, including providing the trial registration number to allow for tracking purposes.*

**** If the publication is a follow-up to an initial publication, please advise.*

18. Identify yet to be published research that may have results available in the near future that could be relevant in the consideration of your application by MSAC (limiting these to the English language only). *Please do not attach full text articles, this is just intended to be a summary.*

	Type of study design*	Title of research (including any trial identifier if relevant)	Short description of research (max 50 words)**	Website link to research (if available)	Date***
1.	Case series	Genetic Study of Familial and Sporadic ALS/Motor Neuron Disease, Miyoshi Myopathy and Other Neuromuscular Disorders NCT01459302, USA	The purpose of this study is to identify additional genes that may cause or put a person at risk for either ALS, other forms of motor neuron disease, Miyoshi Myopathy and Other NMD in the hopes of improving diagnosis and treatment.	https://clinicaltrials.gov/ct2/show/NCT01459302?term=genetic+diagnosis&cond=Neuromuscular+Diseases&draw=2&rank=8	Estimated Study Completion Date October 2018

** Categorise study design, for example meta-analysis, randomised trials, non-randomised trial or observational study, study of diagnostic accuracy, etc.*

***Provide high level information including population numbers and whether patients are being recruited or in post-recruitment.*

****Date of when results will be made available (to the best of your knowledge).*

PART 5 – CLINICAL ENDORSEMENT AND CONSUMER INFORMATION

19. **List all appropriate professional bodies / organisations representing the group(s) of health professionals who provide the service (please attach a statement of clinical relevance from each group nominated):**

    Royal College of Pathologists of Australasia (RCPA)

    Human Genetics Society of Australasia (HGSA)

20. **List any professional bodies / organisations that may be impacted by this medical service (i.e. those who provide the comparator service):**

    It should be noted that the RCPA provides other services used in the diagnostic workup of patients suspected of having a neuromuscular disorder including: histology/immunohistochemistry/Western blot of muscle biopsy samples, testing serum levels of the enzyme, creatine kinase and analysis of the *PMP22* gene for constitutional genetic abnormalities causing peripheral neuropathy.

    Other professional bodies:

    Royal Australasian College of General Practitioners

    Royal Australasian College of Physicians - Paediatrics & Child Health Division

    Australia and New Zealand Association of Neurologists

21. **List the relevant consumer organisations relevant to the proposed medical service (please attach a letter of support for each consumer organisation nominated):**

    Australasian Neuromuscular Network

    Muscular Dystrophy Australia

22. **List the relevant sponsor(s) and / or manufacturer(s) who produce similar products relevant to the proposed medical service:**

    Not applicable

23. **Nominate two experts who could be approached about the proposed medical service and the current clinical management of the service(s):**

    Name of expert 1: **REDACTED**

    Name of expert 2: **REDACTED**

    Name of expert 3: **REDACTED**

    Please note that the Department may also consult with other referrers, proceduralists and disease specialists to obtain their insight.

PART 6 – POPULATION (AND PRIOR TESTS), INTERVENTION, COMPARATOR, OUTCOME (PICO)

PART 6a – INFORMATION ABOUT THE PROPOSED POPULATION

24. Define the medical condition, including providing information on the natural history of the condition and a high-level summary of associated burden of disease in terms of both morbidity and mortality:

Neuromuscular disorders (NMDs) are a broad range of generally progressive disorders affecting the peripheral nervous system, muscle and neuromuscular junctions that present with a high level of clinical and genetic heterogeneity, and overlapping phenotypes (Fattahi et al 2017). The common aspect of all NMD is abnormal muscle function with associated clinical burden. At the most severe end of the spectrum onset can be in utero leading to foetal akinesia, paralysis or reduced movement in utero (Beecroft et al 2018). Paediatric patients may present with early onset symptoms that include a delay in motor milestones, hypotonia, abnormal gait characteristics, frequent falls, respiratory difficulties, and difficulty ascending stairs or arising from the floor. Late onset or adult patients may present with loss of strength, fatigue, episodic weakness, muscle cramps, falls and difficulties with speech and swallowing (McDonald 2012). Many inherited NMDs are multi-systemic, involving cardiac, respiratory, and other organ systems (Kassardjian et al 2016). The significant life-long morbidities of NMD are frequently severely disabling and associated with premature mortality. The clinical and genetic heterogeneities of NMDs make disease diagnosis complicated and expensive, often involving multiple tests (Ankala et al 2015). While historically treatment options for NMD were poor, new developments offer curative interventions or improvements decreasing morbidity and mortality (Dowling et al 2018). However, novel treatments for NMD are guided by the underlying molecular pathology and establishment of a specific genetic diagnosis.

Neuromuscular disorders can be roughly allocated into four categories: muscle disorders such as Duchenne muscular dystrophy (DMD); motor neuron disorders including spinal muscular atrophies (SMAs); neuropathies such as Charcot-Marie-Tooth disease (CMT); and neuromuscular junction disorders (Arnold & Flanigan 2012) It should be noted that genetic heterogeneity exists not only for NMDs as a group but also within the subgroups. For example, there are over 20 genes implicated in autosomal recessive limb-girdle muscular dystrophy (Efthymiou et al 2016). The common features of types of neuromuscular disorders are summarised in

Table 1. They tend to be mostly genetic in origin, can be inherited as autosomal dominant, autosomal recessive, X-linked or mitochondrial traits, however, *de novo* pathogenic variants are relatively common (up to 30% for DMD (Darras et al 2018)) (Laing 2012). Approximately 761 different NMD disorders exist associated with >500 known genes (Fattahi et al 2017) (see Neuromuscular Disorders Gene Table website www.musclegenetable.fr).

Table 1 Common features of hereditary neuromuscular disorders (Arnold & Flanigan 2012)

Neuromuscular disorder	Common features
Myopathies	
Muscular dystrophy	Usually proximal predominant weakness
Distal myopathy	Distal muscle weakness
Congenital myopathy	Early onset with mostly static or slowly progressive weakness
Metabolic myopathy	Slowly progressive or dynamic symptoms of weakness ± exertional rhabdomyolysis and muscle contracture
Mitochondrial myopathies	Weakness ± other systemic features of mitochondrial cytopathy
Muscle channelopathy	Fluctuating symptoms of muscle stiffness (myotonia) or weakness
Motor neuron	
Proximal spinal muscular atrophy	Lower motor neuron dysfunction with proximal muscle weakness
Familial amyotrophic lateral sclerosis	Mixed lower and upper motor neuron dysfunction
Distal spinal muscular atrophy	Distal muscle weakness
Spinobulbar muscular atrophy	Lower motor neuron dysfunction with proximal limb and bulbar weakness
Neuropathy	
Charcot-Marie-Tooth	Distal sensory loss and muscle weakness
Hereditary sensory & autonomic neuropathy	Sensory loss, variable autonomic dysfunction, usually less prominent weakness
Distal hereditary motor neuropathy	Distal weakness, occasional mild sensory loss
Focal/multifocal neuropathy	Recurrent bouts of focal sensory loss and weakness
Nerve channelopathy	Generalised neuropathy with variable features of sensory loss or pain
Neuromuscular junction disorder	
Congenital myasthenic syndromes	Fluctuating weakness

It is impossible to describe the clinical characteristics of all neuromuscular disorders within the confines of this application. The disease burden and epidemiology are presented below in accordance with the major NMD categories.

i. Muscle disorders (myopathies)

Possibly the best characterised myopathies are the muscular dystrophies Duchenne muscular dystrophy (DMD) and Becker muscular dystrophy (BMD). DMD and BMD are X-linked recessive disorders, and therefore occur mostly in males. They are caused by mutations in the *DMD* gene (Flanigan 2014), which encodes dystrophin, a protein primarily located in skeletal and cardiac muscles (NIH 2017). The incidence of DMD is approximately 1:5,000 live births (Ryder et al 2017). DMD presents with delayed motor milestones in early childhood, with a rapid progressive deterioration of muscle tissue and resultant weakness occurring before the teen years. Cardiomyopathy affects almost all DMD patients and is a

common cause of death, usually by 20-30 years of age. BMD is a less common and less severe form of disease than DMD, with a highly variable phenotype characterised by later-onset skeletal muscle weakness, presenting typically between 5-15 years of age. BMD patients tend to live longer than those with DMD, with heart failure from cardiomyopathy being a common cause of death usually in the mid-40s (Darras et al 2018).

Other muscular dystrophies besides dystrophinopathes include:

- Congenital muscular dystrophy: a heterogeneous group of early-onset muscular dystrophies. Affected children are usually symptomatic at birth or in the first 6 months. Congenital muscular dystrophy is characterised by hypotonia, muscle weakness, and reduced deep tendon reflexes, with or without joint contractures. Feeding and respiratory insufficiency are common; additional features may include microcephaly, eye anomalies, cerebral malformation, joint laxity, muscle atrophy, or hypertrophy (Mah et al 2016; Menezes & North 2012).
- Emery-Dreifuss muscular dystrophies are associated with joint contractions, scapulo-peroneal weakness, and cardiac involvement with variable severity and progression, frequently leading to premature mortality (Bonne 2014).
- Facioscapulohumeral muscular dystrophy Type 1 (FSHD1) is a relatively common muscular dystrophy caused by deletion of an exact number of the 3.3kb D4Z4 repeats close to the telomere of the long arm of chromosome 4 (Hewitt et al 1994). Deletion of the D4Z4 repeats associated with FSH1 cannot be detected by NGS, but diagnosis of the phenotypically identical FSHD Type 2 (FSHD2) caused by mutation of *SMCHD1* is detectable by NGS (Lemmers et al 2012). Genetic diagnosis of FSHD1 therefore requires a separate mutation-specific analysis.
- Limb-girdle muscular dystrophies: a heterogeneous group of autosomal muscular dystrophies that may be inherited in a recessive or dominant fashion. Limb girdle muscular dystrophies are characterised by progressive weakness affecting predominantly the hip and shoulder girdles (Mah et al 2016; Menezes & North 2012).

It should be noted that myotonic dystrophy (*DM1*), an autosomal dominant dystrophy associated with clinical myotonia, progressive muscular weakness, and extra-muscular manifestations such as cardiac arrhythmia and endocrine dysfunction, is *not* detected by the genetic panel described in this application, and will therefore not be discussed further. Myotonic dystrophy is caused by the expansion of CTG repeats in the *DMPK* gene and is best diagnosed/detected with the use of triplet primed PCR (Kumara et al 2018).

Congenital myopathies

The congenital myopathies are another large group of muscle disorders, usually presenting at birth, though presentation may be in utero causing foetal akinesia or may be much later in life. Many of the causative genes for the congenital myopathies code for protein components of the sarcomere (Ravenscroft et al 2015).

ii. Motor neuron disorders

Motor neuron disorders include spinal muscular atrophies (SMAs) and amyotrophic lateral sclerosis (ALS), also known as motor neurone disease, or Lou Gehrig's disease. SMA is one of the most common fatal childhood disorders with prevalence in the general population of 1:6,000 to 1:10,000 live births per year (Pearn 1978). SMAs are lower motor neuron disorders affecting the spinal cord anterior horn cells and brain stem motor nuclei. SMA is characterised by progressive proximal weakness, hypotonia, absent tendon reflexes, postural tremor of fingers and tongue fasciculations. The most common form of SMA is an autosomal recessive (AR) disorder (95% of cases), which is caused by a homozygous deletion or mutation of the survival motor neuron 1 (*SMN1*) gene. The classic SMA caused by mutation of *SMN1* overlaps clinically with another AR SMA, spinal muscular atrophy associated with early respiratory failure SMARD1 due to mutations in *IGHMBP2.*

Most of the other SMAs are rare and usually named distal SMA, clinically overlapping with Charcot-Marie-Tooth disease and hereditary spastic paraplegia (Arnold et al 2015; Menezes & North 2012). Genomic

testing cannot detect the classic SMA caused by deletion of *SMN1* due to the variable copy number of *SMN1* and the closely related neighbouring *SMN2*, but can detect the other SMAs.

ALS is characterised by the degeneration of motor neurons in the brain, brainstem and spinal cord that control voluntary and involuntary muscles. Presenting symptoms include limb weakness, weakness of speaking, swallowing and breathing muscles, in addition to muscle spasticity, loss of dexterity and difficulty walking. Most ALS cases are sporadic but 5-10% of cases are familial (Bhatt 2016). The disease ultimately leads to atrophy and impairment of the limb muscles with a proportion of patients progressing to dysphagia, ventilatory compromise, and sialorrhea. Although ALS can affect people of any age, symptoms are usually noted at around 60 years; however, for inherited cases this is usually younger (50 years), with an average survival from onset to death of 2-4 years (Khairoalsindi & Abuzinadah 2018). Juvenile ALS is a very rare severe motor neuron disease that is characterised by progressive upper and lower motor neuron degeneration. Onset of JALS occurs during early childhood at a mean age of 6.5 years (range 3-20 years). Patients develop motor neuron degeneration leading to facial muscle spasticity, spastic dysarthria, and spastic gait, with mild atrophy of the legs and hands observed in some cases (Orphanet 2018).

iii. Peripheral neuropathies

Charcot-Marie-Tooth disease (CMT), which is the most common form of inherited peripheral neuropathy, is the most prevalent hereditary neuromuscular disorder with prevalence of approximately 1:2,500 (Baets et al 2014). CMT is a genetically heterogeneous group of inherited neuropathies, associated with sequence or copy-number variations in over 80 genes coding for proteins with strategic functions in Schwann cell or peripheral axon development and physiology, including myelin proteins, transcription factors, cytoskeletal components, and mitochondrial proteins. In Western countries with mixed ethnicities, autosomal dominant and X-linked dominant forms of CMT disease predominate. More than 90% of CMT disease cases with a molecular diagnosis are associated with pathogenic variants in 4 genes: *PMP22, GJB1, MFN2,* and *MPZ* (Saporta 2014). Severity of symptoms and life expectancy varies widely. CMT is characterised by progressive distal weakness and wasting, a high stepping gait, foot deformities, absent deep tendon reflexes and distal sensory loss, with symptoms usually beginning in early childhood or early adulthood, but can also be late onset (Menezes & North 2012). CMT overlaps clinically with other disorders affecting the peripheral nerve, including some hereditary spastic paraplegias, leukodystrophies and motor neuron disorders, such as juvenile ALS.

iv. Neuromuscular junction disorders

Pathogenic variants in genes encoding proteins which form, regulate or function at the neuromuscular junction are associated with congenital myasthenic syndromes (CMS). CMS usually presents in infancy with a mixture of static and episodic weakness in the facial and limb/girdle muscles. The genetic heterogeneity is underlined by variants in 12 genes, with those in the acetylcholine receptor subunit epsilon (CHRNE) being the most prevalent (Beeson 2016). Different pathogenic variants result in various pathogenetic disease mechanisms where therapeutic options depend on accurate identification of the underlying molecular aetiology (Dowling et al 2018). Some variants in these genes, including the acetylcholine receptor subunit genes result in foetal akinesia (Beecroft et al 2018).

25. Specify any characteristics of patients with the medical condition, or suspected of, who are proposed to be eligible for the proposed medical service, including any details of how a patient would be investigated, managed and referred within the Australian health care system in the lead up to being considered eligible for the service:

Due to the heterogeneous nature of NMDs patients may present with many different clinical features as previously mentioned, with onset occurring at all stages of life from before birth, at birth, early to mid-childhood, right up to late adult onset. Paediatric patients suspected of having an NMD would usually be referred by their general practitioner to a paediatrician, who may then consult with a range of professions including physical therapists, neurologists and cardiologists, in addition to geneticists and genetic counsellors. Patients would undergo numerous diagnostic tests (described below) in order to ascertain a diagnosis including blood tests, physical assessments of gait and movement, muscle biopsies, electromyography (EMG), magnetic resonance imaging (MRI) or nerve conduction studies and in some

cases, cardiological assessments. Adult patients would similarly be referred by their general practitioner to a neurologist.

26. Define and summarise the current clinical management pathway *before* patients would be eligible for the proposed medical service (supplement this summary with an easy to follow flowchart [as an attachment to the Application Form] depicting the current clinical management pathway up to this point):

The diagnosis of neuromuscular disorders traditionally involves taking a detailed patient and family history, conducting a clinical and neurophysiological assessment, followed by a pathological evaluation of muscle and/or nerve biopsy and sequential testing of individual genes (Figure 1**Error! Reference source not found.**) (Teoh et al 2016).

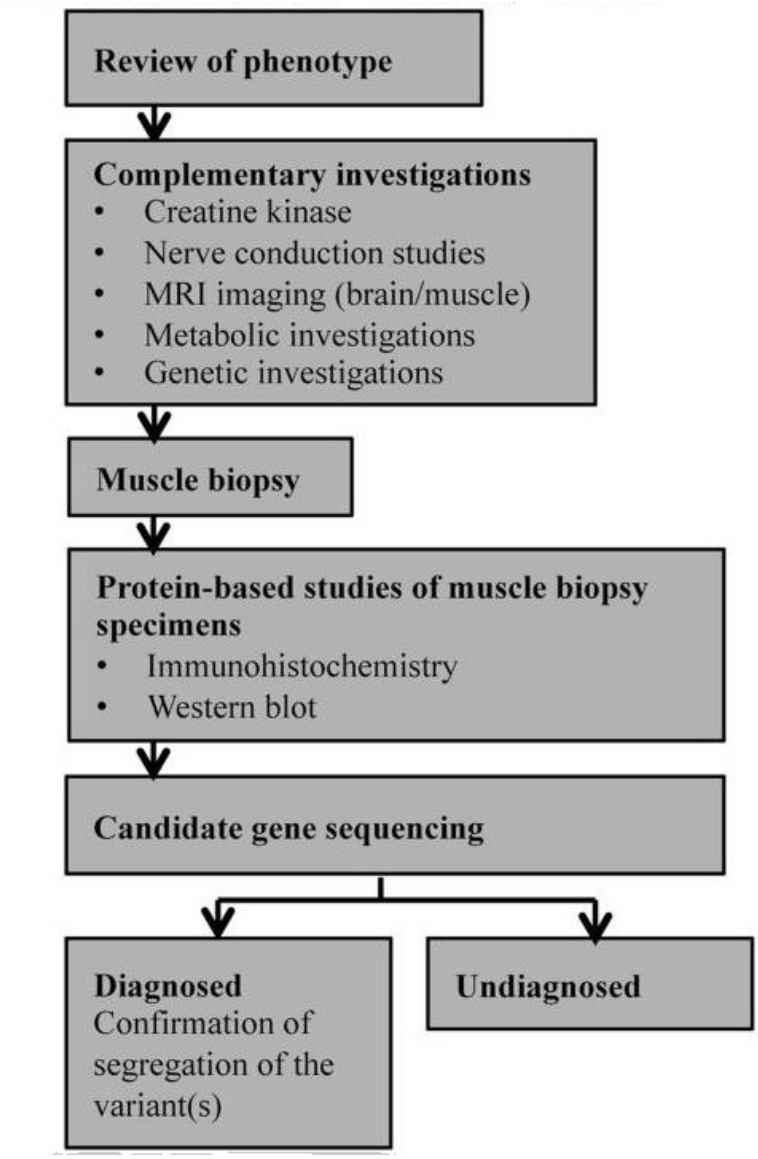

Figure 1 *Conventional diagnostic algorithm based on muscle biopsy and protein-based studies of muscle biopsy specimens, followed by candidate gene sequencing (Schofield et al 2017) Note: complementary investigations would be ordered based on the presenting phenotype of the patient. Not all complementary investigations would be conducted on all patients.*

The congenital myopathies are classified based on structural abnormalities on muscle biopsy, while protein abnormalities on immunohistochemistry and immunoblotting aid classification of the muscular dystrophies (Menezes & North 2012).

A clinical history would include the following details:

- Pregnancy
    - Polyhydramnios, intrauterine movements and contractures
- Birth
    - Congenital hip dislocation, contractures, absent flexion creases
    - Medications given to mother (magnesium sulphate, opioids and anaesthetic agents)
- Neonatal period and infancy
    - Respiratory distress
    - Hypotonia and floppiness
    - Sucking and swallowing difficulties
- Distribution of the weakness
    - Proximal, distal or global
    - Facial or bulbar weakness
    - Ophthalmoplegia
- Family history
    - Consanguinity
    - Family history of a NMD and pedigree (Menezes & North 2012).

A full list of recommended and potentially useful investigative tests is summarised in Table 2.

DMD/BMD: The mean age of patients with suspected DMD/BMD at first clinical presentation/evaluation is 3.6 years, with a mean age at diagnosis of 4.9 years. Creatine kinase (CK) levels are elevated in both DMD and BMD patients due to the increased permeability of the sarcolemmal membrane, therefore patients suspected of DMD or BMD should have CK levels measured. It should be noted that elevated serum CK levels are present at birth in DMD/BMD patients, which offers the opportunity for newborn screening of the disorder (Flanigan 2014). A raised level of creatine kinase should result in referral to a neuromuscular specialist, with input from a geneticist or genetic counsellor. The gold standard of diagnosis (and for differentiating between DMD and BMD) remains muscle biopsy. Biopsy may reveal histological changes such as fibrosis, with immunohistochemical or western blot analysis used to quantify the level, including absence of dystrophin, the protein product of the *DMD* gene (Flanigan 2014). Genetic sequencing has replaced the requirement for initial muscle biopsy. Diagnosis is based on these results combined with the clinical symptoms described above in Q24. Treatment for DMD or BMD would not be commenced until a definitive diagnosis by genetic sequencing (Birnkrant et al 2018b).

Motor neuron disorders, neuropathies and congenital myasthenic syndrome: Serum CK may be normal to moderately raised (up to 5x normal) in SMAs and the congenital myopathies. Electromyography (EMG) and nerve conduction studies may help to differentiate neurogenic from myopathic weakness and to assist in the classification of neuropathy as demyelinating, axonal or intermediate and as sensorimotor or primarily motor or sensory neuropathy. The presence of a decrement in muscle action potential on repetitive nerve stimulation at low rates (2–5 Hz) is useful in identifying those with congenital or acquired myasthenia. Prolonged runs of motor unit potentials with a waxing and waning frequency and amplitude, described as resembling the sound of a motorcycle or chainsaw, may be heard on insertion of the needle electrode on EMG in older patients with myotonic dystrophy. A muscle biopsy may show evidence of chronic denervation, dystrophic change or structural abnormalities. The presence of small angular fibres, fibre-type grouping and type 2 fibre atrophy indicates chronic denervation as seen in SMAs and CMT. On the other hand, in the congenital and limb-girdle dystrophies, the muscle biopsy is characterised by diffuse variation in fibre size, necrotic and regenerating fibres and fibrosis. Nerve biopsy sections and teased fibre preparations are useful in the evaluation of neuropathies that remain genetically unclassified and may show evidence of axonal degeneration or demyelination, helping to more precisely target

genetic studies. Patients with an initial clinical diagnosis of SMA based on history and clinical examination, would undergo first a single gene test for the common *SMN1* deletion, bypassing other investigations including muscle or nerve biopsy (Menezes & North 2012). If found to be negative, these patients would then undergo further genetic testing using an NMD panel to diagnose other SMAs.

Table 2 Essential and recommended investigative tests that would be conducted in the absence of genetic testing for patients suspected of NMD (Schofield et al 2017)

Neonatal/early infantile presentation	Early childhood presentation
Essential investigations	
Blood collection x2	Blood collection x2
Creatine kinase	Creatine kinase
Lactate	Lactate
Ammonia	Ammonia
Urine metabolic screen	Urine metabolic screen
DNA extraction and storage	DNA extraction and storage
Chromosomal microarray	Chromosomal microarray
Investigations that may be considered depending on presentation	
Very long chain fatty acids	Thyroid function studies
Thyroid function studies	Myotonic dystrophy (DM1)
Plasma amino acids	*SMN1* exon copy number (SMA)
Myotonic dystrophy	MRI brain +/- anaesthetic and day stay admission
SMN1 exon copy number (SMA)	Muscle MRI scan +/- anaesthetic and day stay admission
Prader Willi syndrome	Dystrophin MLPA
Acetylcholine receptor antibodies	Ophthalmology review
Antibody screen for congenital infection	Connective tissue dysplasia clinic review
Lumbar puncture with lactate, amino acids and neurotransmitters	
Head ultrasound scan	
MRI brain +/- anaesthetic and day stay admission	
Ophthalmology review	

MLPA = multiplex ligation dependent probe amplification

PART 6b – INFORMATION ABOUT THE INTERVENTION

27. Describe the key components and clinical steps involved in delivering the proposed medical service:

There are four parts to the proposed medical service:

1) Gene panel testing to identify pathogenic variants for genetic neuromuscular disorders in patients where clinical criteria or a family history indicate that genetic testing is warranted.

2) Cascade testing of family members: using mutation-specific detection of a clinically actionable pathogenic variant previously identified in a biological relative.

3) Prenatal genetic testing using mutation-specific detection of a clinically actionable pathogenic variant(s) previously identified in a relative.

4) Prenatal genetic testing for NMD using gene panels: for a suspected NMD in the prenatal period with no previous genetic testing performed in the family, after appropriate counselling

For postnatal diagnostic or cascade testing, the patient would be required to provide a sample containing nucleated cells for DNA extraction (for example, peripheral blood, saliva, buccal swab, etc.). Testing in the prenatal period either due to a known familial cause of NMD or to a clinically suspected NMD with no prior family history requires an amniotic fluid (20ml) or chorionic villi (20mg)[a] sample be obtained.

1) Gene panel testing to identify pathogenic variants for genetic neuromuscular disorders in patients where clinical criteria or a family history indicate that genetic testing is warranted.

The proposed intervention is targeted gene panel analysis of multiple known neuromuscular disease genes using NGS to identify pathogenic variants for genetic neuromuscular disorders in patients where clinical criteria or a family history indicate that genetic testing is warranted.

Over 500 disease genes are currently known for neuromuscular disorders (see Gene Table of Neuromuscular Disorders website www.musclegenetable.fr) and this number is increasing each year as new disease genes are being identified. Known disease genes are being associated with an increased spread of clinical phenotypes (diseases), especially through the use of diagnostic panels or diagnostic exome sequencing (Beecroft et al 2017; Cabrera-Serrano et al 2015). In addition, genes previously associated with only relatively mild dominant disease have been shown to be associated with severe disease through either recessive mutations (Zaharieva et al 2016) or a double dose of a previously presumed exclusive dominant mutation (Kariminejad et al 2017).

As NMDs can be difficult to categorise, a broad panel of genes is considered a better option in order to capture as many causative pathogenic variants as possible. Reducing the number of known disease genes on each panel would reduce the diagnostic yield (Laing 2012).

The proposal is to use two separate panels: a "myopathy" panel for myogenic disorders and a "neuropathy" panel which covers central and peripheral nervous system disorders. Based on clinical criteria, patients will be triaged to undergo testing with *either* the myopathy or neuropathy panel, usually not both; however in some circumstances patients may require testing with both panels.

To maximise the rate of diagnosis, all known myopathy or neuropathy associated genes with mutations detectable by NGS should be included on the panels. The lists of genes included on the current PathWest Laboratory Medicine Neurogenetic Unit myopathy and neuropathy panels are given in Appendix A (318 genes and 375 genes on the myopathy and neuropathy panels, respectively. See Gene Table of Neuromuscular Disorders: www.musclegenetable.fr).

The panels need to be iterated at appropriate intervals with the newly identified disease genes.

Note that some genes are included on both the myopathy and neuropathy panels because variants in some genes can cause what appear to be both myopathic or neuropathic phenotypes. For example, pathogenic variants in *MYH7* encoding slow muscle fibre myosin cause myogenic disorders, including Laing distal myopathy. However, Laing distal myopathy is almost invariably diagnosed clinically as a peripheral neuropathy since that is what it closely resembles and peripheral neuropathy is much more common than distal myopathy (Lamont et al 2014). Another example is variants in *DYNC1H1* which most often cause central or peripheral nervous system disorders but may also result in muscle pathology which can be diagnosed as a congenital myopathy (Beecroft et al 2017).

In addition, it should be noted that whilst most patients will present and be tested at a young age, testing should not be restricted to a paediatric population as many important variants, such as variants in *ACTA1* that encode skeletal muscle alpha actin, may be identified at any stage in development, including adulthood.

Using targeted panels removes some of the issues associated with whole exome analysis, including minimising the risk of incidental findings. In addition, panels are superior to whole exome sequencing or whole genome sequencing for myopathy testing due to the presence of triplicated regions in both *NEB* and *TTN*. In order to

[a] See Victorian Clinical Genetics Services https://www.vcgs.org.au/order/tests/604

identify the pathogenic variants in these triplicated regions it is advantageous to have a *very high depth of coverage* which is easily achieved using targeted panels (Mark Davis et al unpublished data).

Work flow

The proposed workflow for the diagnosis of suspected genetic NMD patients is summarised in Figure 2.

Following referral by a general practitioner to a paediatrician or neurologist, the patient would undergo standard clinical examination to determine if they were candidates for having a genetic NMD.

Any patient suspected of having a genetic NMD would follow one of two pathways:

i) As described in Question 24, the mutations that cause some NMDs, such as facioscapulohumeral muscular dystrophy type 1 (*FSHD1*), myotonic dystrophy type 1 (*DM1*), spinal muscular atrophy type 1 (*SMN1*), are not currently detectable using NGS. Therefore, if there is a strong suspicion on clinical grounds that the patient has one of the disorders not detected by NGS, they should undergo analysis for those single disorders. It should be noted that these individual gene/mutation tests are currently funded either by the patient or by some State health department genetic services as there is no public funding in the form of MBS item numbers for these tests.

If the single disease gene testing is negative, then the patient would undergo analysis using either the myopathic or the neuropathic gene panel, depending on clinical phenotype.

ii) Patients with a suspected NMD other than those that require a separate single gene/mutation test would go straight to testing using either the myopathy or neuropathy gene panel, depending on the clinical phenotype.

2) Cascade genetic testing of family members: for a clinically actionable pathogenic variant(s) previously identified in a biological relative.

Family members of a patient with an NMD where genetic testing has identified the causative variant(s) should receive genetic counselling to establish who is at risk of having the family causative variant(s).

Variant-specific genetic testing, using Sanger sequencing or other technologies such as MLPA, should then be performed for relatives identified as being at risk of having the causative variant(s) identified in the index case.

Testing of parents may be used to establish appropriate segregation of recessive variants identified in the index case, to test whether a pathogenic variant has arisen *de novo* in the index case, or whether a parent is a somatic mosaic for the pathogenic variant. Variant-specific testing may also be used to test affected siblings and for affected relatives in the extended family. It could also be used for pre-symptomatic testing for late-onset disorders in apparently unaffected relatives.

Family members identified as carriers of the family pathogenic variant(s) may then be faced with future reproductive choices to consider, including pre-implantation genetic diagnosis or prenatal genetic testing through chorionic villus or amniotic fluid sampling. For some mutations, carriers may also require clinical assessment and follow-up especially regarding cardiomyopathy.

3) Prenatal genetic testing: **using mutation-specific detection of a clinically actionable pathogenic variant(s) previously identified in a relative**

Prenatal genetic testing for a clinically actionable pathogenic variant(s) should be offered for any future pregnancies after appropriate counselling. The mutation-specific testing would most often use Sanger sequencing to identify the causative pathogenic variant(s) previously identified in the family proband.

4) Prenatal genetic testing for NMD using gene panels: for a suspected NMD in the prenatal period with no previous genetic testing performed in the family, after appropriate counselling.

Where there is a high degree of suspicion that a woman may be carrying a fetus with a NMD e.g. suspected foetal akinesia detected by prenatal ultrasound, consideration should be given to a prenatal gene panel testing in the context of an index patient (fetus).

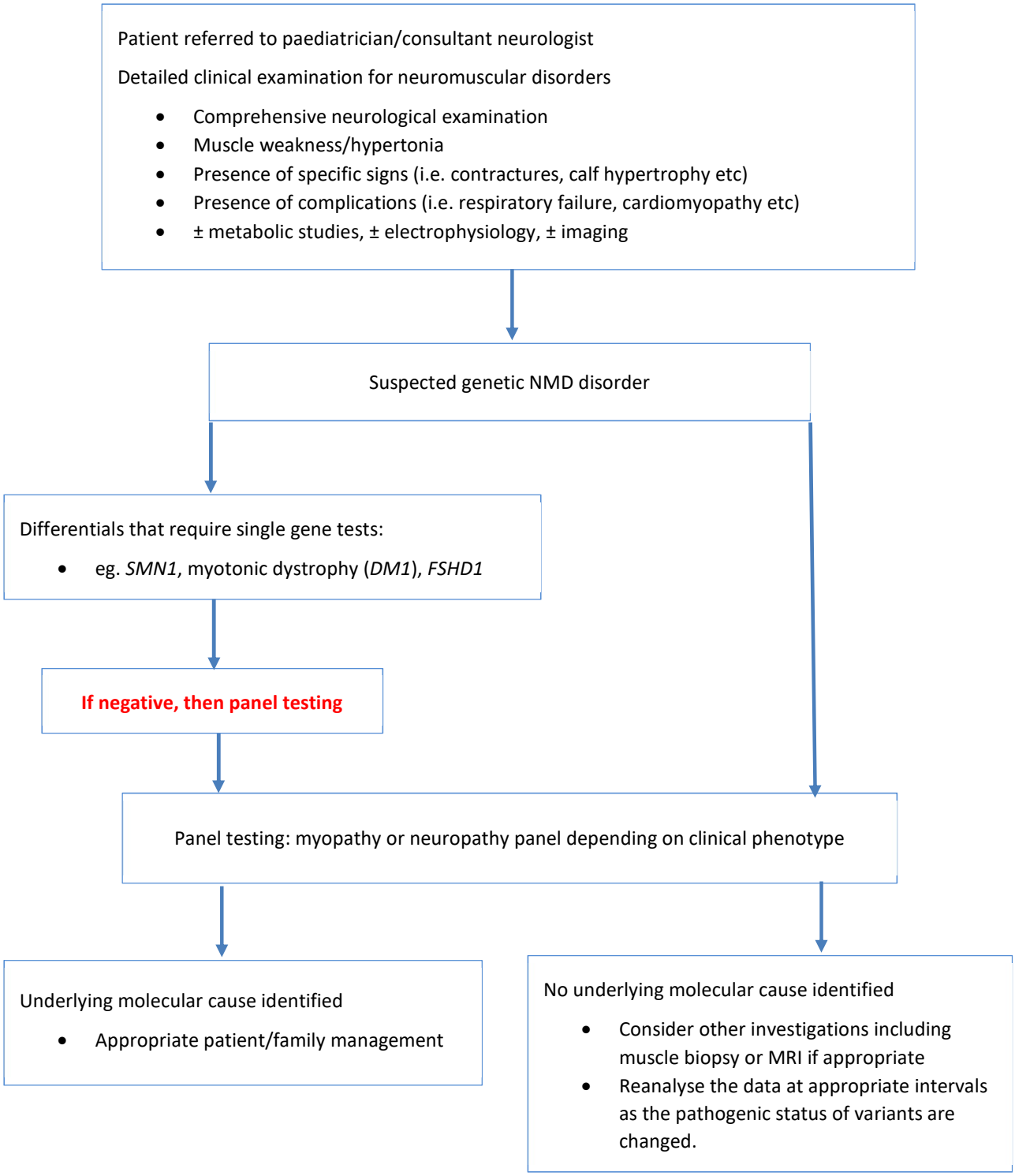

Figure 2 *Proposed molecular approach to diagnosis of NMD patients*
Note: this schematic is a visual aid, depicting the testing pathway; however, it is unlikely that a laboratory would run an individual panel for each disorder e.g. one panel would be used to test for motor neuron, peripheral neuropathies and neuromuscular junction disorders (NMJD)

28. Does the proposed medical service include a registered trademark component with characteristics that distinguishes it from other similar health components?

N/A

29. If the proposed medical service has a prosthesis or device component to it, does it involve a new approach towards managing a particular sub-group of the population with the specific medical condition?

N/A

30. If applicable, are there any limitations on the provision of the proposed medical service delivered to the patient (i.e. accessibility, dosage, quantity, duration or frequency):

One off diagnostic test, noting that patients who test negative may undergo retesting at a future date if the test is expanded to include new genes.

The sequence data may also be reanalysed as the status of variants may change from variants of uncertain significance (VUS) (Class 3) to likely pathogenic (Class 4) or pathogenic (Class 5) with time (Richards et al 2015). Data reanalysis for variant re-classification is formally requested by a clinician if new clinical evidence becomes available in the disease evolution or if clinical re-assessment is aligned with new literature to suggest potential disease causality of a specific, previously unclassified variant.

Data reanalysis may also be prompted by the laboratory in liaison with the requesting clinician if a new gene has been associated with a relevant particular phenotype or if a new clinical association has been reported for a previously known NMD gene. In recent years, WA Health has reported approximately 5 occasions where a new variant was identified after reanalysis under these circumstances.

31. If applicable, identify any healthcare resources or other medical services that would need to be delivered <u>at the same time</u> as the proposed medical service:

Although genetic counselling is a critical component of genomic testing, it is unlikely that counselling would occur prior to, or at the same time as the genetic test due to long waiting lists. The requesting specialist, whether a paediatrician, neurologist or other specialist, may triage the patient to counselling at the same time as requesting the genetic test while explaining the test procedure, benefits and limitations, and potential consequences of test results to the patient/family members. Alternatively, the requesting specialist may triage the patient to counselling after a definitive genetic diagnosis is obtained. Patients may need to be referred to a neurologist for some diagnostic tests, and some may require referral to a cardiologist for diagnosis/treatment of cardiomyopathy.

32. If applicable, advise which health professionals will primarily deliver the proposed service:

Depending on age of onset, a specialist paediatrician, neurologist, or in the case of women considering pregnancy and requiring prenatal testing, a maternal-fetal medicine specialist, would normally request the service.

A pathologist or an appropriately qualified medical scientist would perform the service and provide the clinical report, including interpretation of the results.

33. If applicable, advise whether the proposed medical service could be delegated or referred to another professional for delivery:

N/A

34. If applicable, specify any proposed limitations on who might deliver the proposed medical service, or who might provide a referral for it:

Consideration should be given to restricting those who can provide a referral for this service to a specialised setting: specialist paediatricians, consultant neurologists, clinical geneticists or fetal-maternal specialist.

35. If applicable, advise what type of training or qualifications would be required to perform the proposed service as well as any accreditation requirements to support service delivery:

Testing would be delivered only by NATA Accredited Pathology Laboratories (as defined in MBS Pathology table) by referral only by registered Medical Practitioners (non-pathologists) in line with other tests in the MBS Pathology Table. Interpretation of results would be provided by approved practising pathologists or medical scientists

36. (a) Indicate the proposed setting(s) in which the proposed medical service will be delivered (select all relevant settings):

- ☐ Inpatient private hospital
- ☐ Inpatient public hospital
- ☐ Outpatient clinic
- ☐ Emergency Department
- ☐ Consulting rooms
- ☐ Day surgery centre
- ☐ Residential aged care facility
- ☐ Patient's home
- ☒ Laboratory
- ☐ Other – please specify below

Specify further details here

(b) Where the proposed medical service is provided in more than one setting, please describe the rationale related to each:

N/A

37. Is the proposed medical service intended to be entirely rendered in Australia?

- ☒ Yes
- ☐ No – please specify below

PART 6c – INFORMATION ABOUT THE COMPARATOR(S)

38. Nominate the appropriate comparator(s) for the proposed medical service, i.e. how is the proposed population currently managed in the absence of the proposed medical service being available in the Australian health care system (including identifying health care resources that are needed to be delivered at the same time as the comparator service):

The nominated comparator is no genetic testing.

A number of diagnostic tests, such as serum creatine kinase testing, electro-diagnostic studies including electromyography (EMG) and nerve conduction studies or muscle biopsy can be performed as part of the clinical work up of patients suspected of having an NMD. However, these tests cannot deliver a definitive diagnosis. Muscle biopsy may in some cases provide a strong suggestion of which disease is involved: for example, in Duchenne muscular dystrophy if dystrophin levels are sufficiently reduced (McDonald 2012), however, it cannot identify the precise pathogenic variant in the patient. EMG may define the disease in a patient as a congenital myasthenic syndrome but not which genetic subtype. Therefore these diagnostic tests *cannot* be considered true comparators. Only the identification of the precise disease-causing mutation in the patient in the correct gene can accurately determine which NMD a patient has (Laing 2012).

Therefore, the nominated comparator is no genetic testing.

39. Does the medical service that has been nominated as the comparator have an existing MBS item number(s)?

☐ Yes (please provide all relevant MBS item numbers below)
☒ No

There are MBS item numbers for some of the tests that can be performed to characterise NMDs. There are no MBS item numbers for Sanger sequencing any NMD genes or WEA or WGS for neuromuscular disorders. Muscle biopsy may give an indication of which disease may be involved.

Item number 30075: DIAGNOSTIC BIOPSY OF LYMPH GLAND, MUSCLE OR OTHER DEEP TISSUE OR ORGAN, as an independent procedure, if the biopsy specimen is sent for pathological examination. Fee: $149.75

Item number 72846: Immunohistochemical examination of biopsy material by immunofluorescence, immunoperoxidase or other labelled antibody techniques with multiple antigenic specificities per specimen - 1 to 3 antibodies except those listed in 72848. Fee $59.60

Item number 72844: Enzyme histochemistry of skeletal muscle for investigation of primary degenerative or metabolic muscle diseases or of muscle abnormalities secondary to disease of the central or peripheral nervous system - 1 or more tests. Fee: $30.75

40. Define and summarise the current clinical management pathways that patients may follow *after* they receive the medical service that has been nominated as the comparator (supplement this summary with an easy to follow flowchart [as an attachment to the Application Form] depicting the current clinical management pathway that patients may follow from the point of receiving the comparator onwards including health care resources):

The comparator is no genetic testing. Therefore, the clinical management pathway follows that as described in Figure 1, the conventional care pathway with potential diagnostic tests as described in Q26.

There are multiple issues with trying to summarise all the clinical management pathways NMD patients may follow. The difficulties arise mainly from the spectra of diseases diagnosed by the myopathy and neuropathy panels. One example of a care pathway for a child diagnosed with DMD is given below. However, due to the heterogeneous nature of NMDs it should be noted that the care pathways for all NMDs involve a multi-disciplinary approach, including counselling and referral to a neuromuscular specialist in addition to paediatricians, physiotherapists, respiratory therapists and cardiologists. The clinical care pathways and which services would be required are different for the different categories of NMDs; however, psychosocial care for patients and family members should occur at all stages of disease progression (Birnkrant et al 2018b; Birnkrant et al 2018c).

DMD: Early stage management of DMD while the child is still ambulatory includes:

- Physiotherapy for advice on stretching, to prevent contractures
    - Later, knee-foot-ankle orthoses may help prolong walking
    - Serial casting of the ankles may be helpful (may prevent the need for surgical release of the Achilles tendon).
- Glucocorticoid therapy, which may prolong ambulation by 6-24 months, preserve upper limb and respiratory function, and avoid the need for scoliosis surgery
    - note of caution as these may be associated with side-effects including osteoporosis and vertebral fractures
    - Prednisone or prednisolone 0·75 mg/kg per day is the usual treatment, or deflazacort 0·9 mg/kg per day
- Optimisation of bone health
    - vitamin D and calcium dietary advice or supplements.
    - bisphosphonates if vertebral fracture occurs (Birnkrant et al 2018b).

Once mobility is lost:

- Continue steroid use but reduce dose as necessary to manage side-effects
- Help with mobility - usually an electric wheelchair
- Orthopaedic care - orthotics or surgery for contractures and scoliosis. Scoliosis is usually progressive and treated with surgery
- Cardiac and respiratory surveillance
    - physiotherapy
    - non-invasive ventilation to relieve symptoms and prolong survival, may include assist coughing and airway clearance by manual techniques, cough assist devices or tracheostomy
    - ECG and echocardiograms may be difficult to interpret due to scoliosis
    - Cardiac impairment and arrhythmias treated with ACE inhibitors, diuretics and beta-blockers
    - treat nocturnal hypoventilation
    - increase cardiac monitoring if treating with glucocorticoids, noting weight gain and blood pressure
    - patients are at increased risk of thromboembolism; consider anticoagulation if there is severe cardiac impairment.
- Palliative care (Birnkrant et al 2018a).

Female carriers of a disease-causing DMD mutation are at risk of skeletal muscle disease and cardiomyopathy. A baseline cardiac assessment in early adulthood that includes an electrocardiogram and non-invasive imaging, preferably cardiovascular MRI is recommended. Ongoing surveillance is required, with guidelines suggesting assessment every 3–5 years (Birnkrant et al 2018a). Female carriers should also be offered family planning options including pre-implantation genetic diagnosis or prenatal genetic testing through chorionic villus or amniotic fluid sampling for male pregnancies.

41. (a) Will the proposed medical service be used in addition to, or instead of, the nominated comparator(s)?

☐ In addition to
☒ Instead of

(b) If instead of please outline the extent of which the current service/comparator is expected to be substituted:

All patients presenting with a clinical suspicion of an NMD should be referred on for mutational analysis after initial clinical tests. This would negate the need to perform an invasive and painful skeletal muscle biopsy in all cases for whom a definitive molecular diagnosis is obtained. Only those cases that return a negative genetic testing result should undergo a muscle biopsy.

42. Define and summarise how current clinical management pathways (from the point of service delivery onwards) are expected to change as a consequence of introducing the proposed medical service including variation in health care resources (Refer to Question 39 as baseline):

Genetic testing plays a role in the diagnosis, appropriate investigation, and monitoring of NMDs. A definitive diagnosis by mutational analysis will negate the need to perform many other downstream diagnostic tests, including invasive procedures such as muscle or nerve biopsies (and the associated Western blot or immunohistochemistry), which carry a small but recognised morbidity (McDonald 2012). Other expensive testing may also be avoided, including nerve or muscle imaging, and blood and cerebrospinal fluid testing. In determining the index case, additional testing of other family members for other disorders may be avoided. In the absence of gene panel testing, patients may undergo time consuming and expensive sequential Sanger sequencing (so called "diagnosis by sequencing" and not MBS listed) of the most likely candidate genes in relation to the suspected clinical phenotype in a bid to confirm a diagnosis. This, as described above (Q 38) is not feasible for all of the now identified NMD genes.

Panel testing maximises the chance of identifying the causative variant(s) in a patient, with a genetic diagnosis potentially changing the clinical diagnosis (Beecroft et al 2017; Leidenroth et al 2012) and the likelihood that a treatable disease might be identified. By reaching a timelier diagnosis with gene panel testing, appropriate treatment of patients may commence earlier for the NMDs for which effective therapies are available. In addition, by arriving at a definitive molecular diagnosis, appropriate and potentially life-saving surveillance or referrals may be commenced, especially regarding those disorders with a cardiac or respiratory component. For example:

- Patients diagnosed as DMD will commence treatment with steroids immediately, which may prolong the time that they are ambulatory and improve their quality of life.
- New treatments in development for DMD patients include antisense oligonucleotide exon skipping drugs aim to restore the disrupted reading frame allowing the production of truncated but partly functional dystrophin proteins, slowing progression of the disease (Shimo et al 2018).
- Other pharmaceutical options are available for some patients include ion channel therapeutic drugs available to treat patients with myotonias disorder and treatments for congenital myasthenic syndromes that are gene-specific. For example, identifying mutations in DOK7 in patients indicates treatment with Salbutamol should be instigated; however, other myasthenias can be treated with mestinon, which in some patients can stop life-threatening admissions to ICU with chest infections (personal communication WA Health).
- Genomic diagnosis is also useful in Pompe disease, which is difficult to diagnose in adult patients, as it then allows for treatment with intravenous enzyme replacement therapy.
- Titin, encoded by the gene TTN, is the largest human protein, and plays central roles in sarcomeric structures and functions in skeletal and cardiac muscles. Specific mutations in TTN are causally related to specific types of muscular dystrophies and cardiomyopathies.(Misaka et al 2019; Yoshihisa et al 2018) Prior to genomic diagnosis, it was not possible to analyse these mutations because of the size and repetitive nature of the gene. With genomic diagnosis sudden cardiac death due to dilated cardiomyopathy can be prevented (personal communication WA Health).
- Similarly, diagnosis of mutations in LMNA (Lamin A/C), a neuromuscular junction defect allows close monitoring of cardiac rhythm to prevent sudden cardiac death (personal communication WA Health).
- Referral to a cardiologist for investigation and consideration of implantation of a defibrillator or pacemaker in a patient with limb girdle muscular dystrophy.
- Quality of life may also be improved with a definitive diagnosis that may prompt screening for treatable comorbidities such as cataracts or diabetes in a patient with myotonic muscular dystrophy.

- Detecting mutations in the *ABCD1* gene indicates that Addison's disease should be considered, which may be potentially life-saving (personal communication WA Health).
- Identification of an AT mutation in an atypical adult ataxia telangiectasia patient indicates breast cancer screening should be investigated. In one family this led to the detection of early breast cancer in both the 25 year-old patient and her carrier mother. Without a genetic diagnosis, clinical indicators alone would not have led to this diagnosis in a patient with no telangiectasia (personal communication WA Health).

It should be noted; however, that many of these interventions are variant-specific. Treatment of the wrong variant with the wrong drug will result in a worsening of the disorder.

There is clinical utility for family members to know about an inherited disorder, because the *specific genetic diagnosis* helps to predict the pattern of inheritance. This information is critical for genetic counselling, specifically informing family planning discussions, as well as discussions around prenatal diagnosis or early diagnosis of asymptomatic family members, as well as providing patient access to specific disease support groups. Without a genetic diagnosis, physicians or genetic counsellors cannot adequately educate patients or predict risks in a manner that helps guide family planning or diagnostic decisions (Kassardjian et al 2016).

PART 6d – INFORMATION ABOUT THE CLINICAL OUTCOME

43. Summarise the clinical claims for the proposed medical service against the appropriate comparator(s), in terms of consequences for health outcomes (comparative benefits and harms):

Public funding of genetic testing for the diagnosis of NMDs would provide equity of access. Currently some patients are accessing genomic testing for NMDs on a user-pays basis by sending samples overseas to Invitae[b] at a cost of up to US$1,500. It should be noted that this panel only contains 109 genes, and therefore may leave many patients without a diagnosis. Importantly, this testing is not covered by the same quality assurance processes as Australian NATA accredited laboratories are subject to and there have been poor outcomes with low "hit" rates reported. NMD gene panel testing is currently tested in a Centre of Excellence in Western Australia.

Providing a definitive diagnosis gives certainty to patients and families. The importance of being able to give the family disease a name through accurate molecular diagnosis cannot be overemphasised. The family now knows what disease they have and knowing what disease they have provides closure (Basel & McCarrier 2017). It stops the family being in limbo. It stops them being in the dark as to what their disease is. It provides clarity as to the pattern of inheritance and therefore clarity of prognosis and clarity as to the risk of a couple having further affected children.

Having an accurate molecular diagnosis also restores reproductive confidence. One example of this that we have written about was when the deletions of *SMN1* responsible for spinal muscular atrophy were first identified in 1995 (Lefebvre et al 1995). When the WA laboratory could first perform prenatal diagnosis for the deletions, we were inundated with an avalanche of WA families wanting prenatal diagnosis, families who had put off having any further children until their disease could be tested for (Laing 2012). The advantage of panel testing is that it maximises the chance of identifying the causative variants for a family, including being able to interpret the triplicated regions of the giant genes *NEB* and *TTN*. Panel testing therefore maximises the chances of being able to restore reproductive confidence for NMD families.

Gene panel testing for NMDs markedly reduces the time to diagnosis compared to standard treatment, which was reported recently, based on Australian neuromuscular centre data, as a mean of 7.7 years (range 2 months to 26 years) (Schofield et al 2017).

The main consequences of introducing genetic testing for the diagnosis of NMDs include:

- Early identification and early commencement of treatment of affected probands, resulting in an improved quality of life and an increased survival time;
- Early identification and early commencement of treatment of affected relatives; and
- Carriers would be positively identified and offered surveillance and family planning options for any future pregnancies.

44. Please advise if the overall clinical claim is for:

☒ Superiority
☐ Non-inferiority

[b] https://www.invitae.com/en/physician/tests/03280/

45. Below, list the key health outcomes (major and minor – prioritising major key health outcomes first) that will need to be specifically measured in assessing the clinical claim of the proposed medical service versus the comparator:

Direct evidence:

Safety Outcomes: Adverse events from testing: physical and/or psychological harms from genetic testing

Clinical Effectiveness Outcomes: Quality of life, morbidity, increased survival time

Cost-effectiveness

Indirect evidence:

Analytical validity: test failure rate, sensitivity, specificity, concordance, unsatisfactory or uninterpretable results, diagnostic yield

Clinical validity: predictive or prognostic value

Therapeutic efficacy: time to diagnosis, change in patient management (earlier access to treatment), change in detection and treatment of family members (earlier detection of and management of relatives due to genetic testing, impact on family planning)

Therapeutic effectiveness: effect of change in management (e.g. increased time to mortality, improved quality of life).

PART 7 – INFORMATION ABOUT ESTIMATED UTILISATION

46. Estimate the prevalence and/or incidence of the proposed population:

Australian data on the incidence and prevalence of NMDs are lacking.

The cross-sectional analysis by Norwood et al (2009) was conducted in the northern counties of England[c] and may therefore be generalisable to Australia. However, prevalence studies are likely to underestimate those diseases that lead to early death, such as DMD, as prevalence reflects both incidence and duration. Norwood et al included all registered patients with genetic muscle diseases diagnosed and currently seen by the neuromuscular team at the UK's Institute of Human Genetics. Patients with inherited disorders such as mitochondrial and metabolic myopathies, ion channel disorders, congenital myasthenic syndromes and hereditary motor and sensory neuropathies as well as those with acquired neuromuscular disease including inflammatory myopathies, myasthenia gravis and motor neuron diseases were excluded. A total of 1,105 patients were identified for inclusion, representing 31 different disease entities. Diagnostic clarity achieved through careful delineation of clinical features supported by histological, immunological and genetic analysis enabled a definitive diagnosis in 75.7% of patients (Norwood et al 2009).

Despite the exclusion of common groups such as hereditary motor and sensory neuropathy (40/100,000) and mitochondrial disorders (9.2/100,000), the combined prevalence was 37.0/100,000. Following that group were patients diagnosed with the dystrophinopathies making up 22.9% (8.46/100,000) of the clinic population. A point prevalence of 7.29 (95% CI [5.98, 8.79]) and 8.29 (95% CI [6.90, 9.88]) per 100,000 males was reported for BMD and DMD, respectively. Limb girdle muscular dystrophy, comprised 6.2% of the clinic population with a combined prevalence of 2.27/100 000. In addition, patients were diagnosed with 12 other muscle disorders, which ranged from a point prevalence of 0.89/100,000 for the group of congenital muscular dystrophies, to conditions with only two affected individuals in a population of three million. The X-linked form of Emery–Dreifuss muscular dystrophy and Ullrich muscular dystrophy had a prevalence of 0.13/100 000, making both very rare. Bethlem myopathy was relatively more common with a prevalence of 0.77/100 000 (Norwood et al 2009).

Incidence

There is a paucity of data on the incidence of NMDs in Australia. In 2015, the Western Australian registry of developmental anomalies reported the incidence of "muscular dystrophies and myopathy" as approximately 0.2/1,000 or 1/5,000 live births,(Bhatt 2016) which is similar to estimates for the incidence of Duchenne muscular dystrophy (1 in 3,500–5,000 male live births) (Matthews et al 2016). Using the number of live births in Australia to estimate the size of the at-risk population would yield an approximate incidence of 62 affected live born individuals each year, based on the 311,104 births registered in 2016. When compared to world-wide incidence data, this estimate is very close to the incidence of DMD, and therefore likely to be an underestimation of the true incidence of NMDs in Australia. In addition, the true incidence is likely to be higher due to the number of (currently) undiagnosed cases.

Two recent systematic reviews have estimated incidence rates for 13 neuromuscular disorders (see Table 3), which, as individual disorders, are rare, but as a group they are not, with an overall incidence rate ranging from 11.95 to 82.8 per 100,000 (Bhatt 2016; Deenen et al 2015). Similarly, this is an underestimation due to the number of NMDs without incidence data.

[c] Northumberland, Durham, Cumbria and parts of Yorkshire and Lancashire with an estimated total population of 2.99 million

Table 3 Global incidence rates for 13 neuromuscular disorders (Bhatt 2016; Deenen et al 2015)

Disorder	Incidence range per 100,000
Anterior horn cells	
Type I spinal muscular atrophy	3.53 to 9.8
All spinal muscular atrophy	3.53 to 14.9
Amyotrophic lateral sclerosis	0.42 to 5.3
Peripheral nerve	
Chronic inflammatory demyelinating polyneuropathy	0.35 to 1.6
Guillain-Bare syndrome	0.4 to 3.0
Friedreich Ataxia	2.7 to 6.19
Neuromuscular junction	
Myasthenia Gravis	0.3 to 2.8 (11.8 reported by one study in Japan)
Lambert-Eaton myasthenic syndrome	0.05
Muscular	
Duchene muscular dystrophy	2.0 to 34.7 per 100,000 males
Becker muscular dystrophy	1.06 to 7.2 per 100,000 males
Limb-girdle muscular dystrophy	0.7
Polymyositis	0.27 to 3.80
Dermatomyositis	0.08 to 1.78
Inclusion body myositis	0.09 to 0.79
TOTAL	**11.95 to 82.8**

47. Estimate the number of times the proposed medical service(s) would be delivered to a patient per year:

Once in a lifetime diagnostic test if a definitive diagnosis is obtained.

48. How many years would the proposed medical service(s) be required for the patient?

N/A

49. Estimate the projected number of patients who will utilise the proposed medical service(s) for the first full year:

Genetic testing for NMDs is currently predominantly conducted in Western Australia for patients from Queensland, NSW, South Australia, Victoria and Western Australia. Although the number of tests conducted in WA Is not inclusive of all NMD patients in Australia, it does represent a significant majority. In addition, a number of prevalent, undiagnosed patients may also seek to use the proposed service.

Data from the previous two years (2017 and 2018) of testing patients with a suspected NMD with an NGS panel are summarised below. The number of cascade tests conducted is dependent on the number of index cases found to be positive. In 2017 and 2018, 29% and 23.7% of patients suspected of having an

NMD were found to be positive for a NMD identified by the NGS panel, respectively. These numbers resulted in 1.12 and 1.3, respectively, family members per index case seeking cascade testing.

The expected numbers for the following 3-years were calculated using the slight increase in the number of tests performed from 2017 to 2018 (an increase of 8.4%). The estimated number of cascade tests performed will be based on the number of positive index cases – an average of the number of positives was used to estimate this (26.35%). Similarly, the estimated number of cascade tests used the average number of tests performed per index case (1.21). It is difficult to estimate the number of prenatal diagnoses, however, the numbers are expected to remain low.

It is expected that uptake of testing should remain relatively constant over the next three years.

Table 4 Number of tests conducted in Western Australia in previous 2 years, and expected number of tests for the next 3 years

Population to be tested	2017	2018	Expected 2019	Expected 2020	Expected 2021
Index cases	944 276 +ve	1023 242 +ve	1108 293	1200 316	1300 342
Cascade testing	310	330	354	382	413
Prenatal diagnosis	13	19			

50. Estimate the anticipated uptake of the proposed medical service over the next three years factoring in any constraints in the health system in meeting the needs of the proposed population (such as supply and demand factors) as well as provide commentary on risk of 'leakage' to populations not targeted by the service:

There are four parts to the proposed medical service:

1) Gene panel testing to identify pathogenic variants for genetic neuromuscular disorders in patients where clinical criteria or a family history indicate that genetic testing is warranted.

2) Cascade testing of family members: using mutation-specific detection of a clinically actionable pathogenic variant previously identified in a first-degree relative.

3) Prenatal genetic testing using mutation-specific detection of a clinically actionable pathogenic variant(s) previously identified in a relative.

4) Prenatal genetic testing for NMD using gene panels: for a suspected NMD in the prenatal period with no previous genetic testing performed in the family, after appropriate counselling.

There is minimal risk of leakage associated with items 1 and 4; however, items 2 and 3 may result in item numbers for cascade testing and prenatal testing of relatives of any proband with a causal variant or variants identified by any technique, including single gene/mutation detection. The item number should specify that the gene panel must be the testing method that was used to identify the familial variant.

PART 8 – COST INFORMATION

51. Indicate the likely cost of providing the proposed medical service. Where possible, please provide overall cost and breakdown:

Cost of index case testing: $1,200

Cost of variant-specific cascade testing: $450

The cost of prenatal testing for a documented familial gene variant, including sampling cost followed by genetic analysis, and maternal cell contamination: $1,000

The cost of prenatal testing, including sampling cost followed by genetic analysis, and maternal cell contamination: $ 1,600

Note: The scheduled fees (100%) stated in the item number descriptors below reflect current costs to perform these analyses. If the scheduled fees are set at these prices, then an 85% reimbursement would result in the need for considerable out-of-pocket payments by patients, with the potential for a lack of equity in access for some patients/families.

In addition to the costs outlined above, prenatal genetic testing would utilise either:

- Chorionic villus sampling, usually after 11 weeks into the pregnancy.
  MBS item number 16603: CHORIONIC VILLUS SAMPLING, by any route.
  Fee: $121.85 Benefit: 75% = $91.40 85% = $103.60;
- Amniocentesis, which isn't usually carried out until 15 to 16 weeks of pregnancy
  MBS item number 16606: Fetal blood sampling, using interventional techniques from umbilical cord or fetus, including fetal neuromuscular blockade and amniocentesis, (Anaes.)
  Fee: $243.25 Benefit: 75% = $182.45 85% = $206.80

The cost of pre-implantation genetic diagnosis may also be considered (MSAC application 1165).

52. Specify how long the proposed medical service typically takes to perform:

Diagnostic testing would take between 4 - 12 weeks, with carrier testing taking 4 - 6 weeks.

Due to workload commitments, the normal turnaround time for testing is 2-4 months (90% of tests requested would fall into this timeline), however urgent cases can be processed in 1-2 weeks.

Prenatal testing for known familial variant(s) 4 -7 days.

53. If public funding is sought through the MBS, please draft a proposed MBS item descriptor to define the population and medical service usage characteristics that would define eligibility for MBS funding.

Item number AAAA	Category 6 (Pathology Services) – Group P7 Genetics
Characterisation of a germline gene variant(s) by a gene panel in a patient presenting with clinical symptoms suggestive of a genetic neuromuscular disorder other than those associated with variants that are not detected by massively parallel sequencing, and after exclusion of non-genetic causes, requested by a specialist or consultant physician. **Fee**: $ 1,200 **Benefit:** 75% = 825.00 85% = 935.00	

Item number BBBB	Category 6 (Pathology Services) – Group P7 Genetics
Detection of a single identified gene variant in a biological relative of a patient with a documented pathogenic germline gene variant for a neuromuscular disorder identified by item number AAAA, requested by a specialist or consultant physician who manages the treatment of the patient. **Fee:** $ 450.00 **Benefit:** 75% = 337.50 85% = 382.50	

Item number CCCC	Category 6 (Pathology Services) – Group P7 Genetics
Prenatal detection of a familial gene variant(s) for a neuromuscular disorder previously identified in an index patient in the family by item number AAAA, including maternal cell contamination assessment, requested by a specialist or consultant physician who manages the treatment of the patient. **Fee:** $ 1,000.00 **Benefit:** 75% = $750.00 85% =$ 850.00	

Item number DDDD	Category 6 (Pathology Services) – Group P7 Genetics
Prenatal detection of an unknown germline gene variant(s) for a suspected genetic neuromuscular disorder using a gene panel, after exclusion of non-genetic causes, and including maternal cell contamination assessment, requested by a specialist or consultant physician who manages the treatment of the patient. **Fee:** $ 1,600.00 **Benefit:** 75% = $750.00 85% =$ 850.00	

Appendix A: Muscular panel

AARS2	*AMPD1*	*C8orf22*	*CHRND*	*COL6A3*	*DPAGT1*	*FHL1*	*GYG1*	*ISPD*	*LDB3*	*MPZ*	*MYL12A*	*NUP88*	*PIEZO2*	*PRELP*
ABHD5	*ANKRD1*	*CA3*	*CHRNE*	*COLQ*	*DPM1*	*FHL3*	*GYS1*	*ITGA7*	*LDHA*	*MRPL3*	*MYL2*	*OBSCN*	*PIGY*	*PREPL*
ACAD9	*ANKRD2*	*CACNA1S*	*CHRNG*	*COX14*	*DPM2*	*FKBP14*	*HACD1*	*ITGA9*	*LIMS2*	*MSTN*	*MYL3*	*OPA1*	*PIP5K1C*	*PRX*
ACADVL	*ANO5*	*CAPN3*	*CHST14*	*COX6A2*	*DPM3*	*FKRP*	*HADHA*	*KBTBD13*	*LMNA*	*MTM1*	*MYL4*	*ORAI1*	*PLEC*	*PTRF*
ACTA1	*APOD*	*CASQ1*	*CKM*	*COX6B1*	*DYSF*	*FKTN*	*HADHB*	*KCNA1*	*LMOD2*	*MTMR14*	*MYLPF*	*P4HA1*	*PLOD1*	*PYGM*
ACTC1	*ASCC1*	*CAV3*	*CKMT2*	*CPT2*	*ECEL1*	*FLNA*	*HBB*	*KCNJ2*	*LMOD3*	*MUSK*	*MYO18B*	*PABPN1*	*PMP22*	*PYROXD1*
ACTN2	*ATP1A2*	*CCDC78*	*CLCN1*	*CRYAB*	*EDN3*	*FLNC*	*HIST1H3H*	*KIF21A*	*LPIN1*	*MUSTN1*	*MYOD1*	*PBLD*	*PNPLA2*	*RAPSN*
ADCY6	*ATP2A1*	*CD99*	*CLDN11*	*CYC1*	*EGR2*	*GAA*	*HNRNPA1*	*KLHL31*	*LRP4*	*MYBPC1*	*MYOT*	*PDE4DIP*	*POGLUT1*	*RBCK1*
ADGRG6	*ATP5A1*	*CEP89*	*CMYA5*	*DAG1*	*EMD*	*GAPDH*	*HNRNPA2B1*	*KLHL40*	*LYZ*	*MYBPC3*	*MYOZ1*	*PDK4*	*POLG*	*RMND1*
ADSSL1	*B3GALNT2*	*CFL2*	*CNTN1*	*DCST2*	*ENO3*	*GBE1*	*HNRNPDL*	*KLHL41*	*MAGEL2*	*MYF6*	*NALCN*	*PDLIM3*	*POLG2*	*RRM2B*
AGK	*B3GALT6*	*CHAT*	*CNTNAP1*	*DES*	*ERBB3*	*GFPT1*	*HRAS*	*KLHL9*	*MB*	*MYH13*	*NDUFAF1*	*PFKM*	*POMGNT1*	*RYR1*
AGL	*B4GAT1*	*CHCHD10*	*COL12A1*	*DLK1*	*ETFDH*	*GGPS1*	*HSPB6*	*KY*	*MEG3*	*MYH14*	*NDUFAF2*	*PGAM2*	*POMGNT2*	*SCN4A*
AGRN	*BAG3*	*CHD7*	*COL13A1*	*DMD*	*FARS2*	*GLE1*	*HSPB7*	*L1CAM*	*MEGF10*	*MYH2*	*NEB*	*PGK1*	*POMK*	*SDHA*
AIFM1	*BIN1*	*CHKA*	*COL1A1*	*DNA2*	*FBN2*	*GMPPB*	*HSPB8*	*LAMA2*	*MGME1*	*MYH3*	*NEFL*	*PGM1*	*POMT1*	*SEPN1*
ALDOA	*BVES*	*CHKB*	*COL3A1*	*DNAJB6*	*FBXL4*	*GNE*	*HSPG2*	*LAMB2*	*MICU1*	*MYH7*	*NFU1*	*PHKA1*	*POMT2*	*SGCA*
ALG14	*C10orf2*	*CHRNA1*	*COL6A1*	*DNM2*	*FDX1L*	*GOLGA2*	*IGF2*	*LAMP2*	*MMP1*	*MYH8*	*NNAT*	*PHKB*	*POSTN*	*SGCB*
ALG2	*C12orf65*	*CHRNB1*	*COL6A2*	*DOK7*	*FGF7*	*GREM1*	*ISCU*	*LARGE*	*MMP28*	*MYL1*	*NRAP*	*PHOX2A*	*PPP1R27*	*SGCD*

SGCG	*SLC25A4*	*SMCHD1*	*SRPK3*	*SURF1*	*TANGO2*	*TIA1*	*TNNC2*	*TNNT3*	*TPM2*	*TRIP4*	*UNC80*	*XIRP2*
SLC18A3	*SLC25A42*	*SNAP25*	*STAC3*	*SYNE1*	*TAZ*	*TK2*	*TNNI1*	*TNPO3*	*TPM3*	*TRPV4*	*UQCRC2*	*ZBTB42*
SLC19A3	*SLC25A6*	*SOX10*	*STIM1*	*SYNE2*	*TBCK*	*TMEM43*	*TNNI2*	*TNXB*	*TRAPPC11*	*TTN*	*VCP*	*ZC4H2*
SLC22A5	*SLC28A2*	*SPEG*	*STMN2*	*SYNPO2*	*TCAP*	*TMEM5*	*TNNT1*	*TOR1AIP1*	*TRIM32*	*UBA1*	*VMA21*	
SLC25A20	*SLN*	*SQSTM1*	*SUCLA2*	*SYT2*	*TGFB3*	*TNNC1*	*TNNT2*	*TP63*	*TRIM63*	*UNC79*	*XIRP1*	

Neuropathy panel

AARS	*ARFGEF2*	*C19orf12*	*DCX*	*EXOSC8*	*GFAP*	*IKBKAP*	*LYST*	*NTRK1*	*POLR3B*	*RTN2*	*SLC52A2*	*TBR1*	*UBA1*
ABCB7	*ARHGEF10*	*CACNA1A*	*DDHD1*	*FA2H*	*GJB1*	*INF2*	*MAG*	*OPA1*	*POMGNT1*	*RTTN*	*SLC52A3*	*TDP1*	*UBA5*
ABCD1	*ARL6IP1*	*CACNA1B*	*DDHD2*	*FAM126A*	*GJB3*	*ISPD*	*MARS*	*OPTN*	*POMGNT2*	*RUBCN*	*SLC5A7*	*TECPR2*	*UBQLN2*
ABHD12	*ARSA*	*CACNA1G*	*DENR*	*FAM134B*	*GJC2*	*ITPR1*	*MARS2*	*OTUD4*	*POMK*	*SACS*	*SLC6A3*	*TFG*	*UNC79*
ACTB	*ARSI*	*CACNB4*	*DHTKD1*	*FARS2*	*GNAL*	*KARS*	*MATR3*	*PAFAH1B1*	*POMT1*	*SAMHD1*	*SLC9A1*	*TH*	*UNC80*
ACTG1	*ARX*	*CCT5*	*DNAJB2*	*FAT3*	*GNB4*	*KCNA1*	*MCPH1*	*PANK2*	*POMT2*	*SBF1*	*SNX14*	*THAP1*	*USP8*
ADAR	*ASAH1*	*CDK5RAP2*	*DNAJB5*	*FAT4*	*GOSR2*	*KCNC3*	*MED25*	*PARK2*	*PPP2R2B*	*SBF2*	*SOD1*	*TNFAIP1*	*VAMP1*
ADCK3	*ASCC1*	*CENPJ*	*DNM2*	*FBLN5*	*GRID2*	*KCND3*	*MFN2*	*PAX6*	*PRDM12*	*SCN11A*	*SOX10*	*TOR1A*	*VAPB*
ADGRG1	*ASPA*	*CEP152*	*DNMT1*	*FBXO38*	*GRM1*	*KCTD13*	*MLC1*	*PBLD*	*PRKCG*	*SCN1A*	*SPAST*	*TOR1AIP1*	*VCP*
AFG3L2	*ASPM*	*CHCHD10*	*DRD2*	*FGD4*	*HACE1*	*KIAA0196*	*MMP28*	*PCNT*	*PRKRA*	*SCN9A*	*SPG11*	*TP63*	*VLDLR*
AIFM1	*ATCAY*	*CHMP2B*	*DRP2*	*FGF14*	*HARS*	*KIF1A*	*MORC2*	*PDK3*	*PRNP*	*SCYL1*	*SPG20*	*TPP1*	*VPS13A*
AIMP1	*ATL1*	*CLCN2*	*DST*	*FIG4*	*HEPACAM*	*KIF1B*	*MPZ*	*PDYN*	*PRPH*	*SEPT9*	*SPG21*	*TREX1*	*VPS37A*

AARS	*ARFGEF2*	*C19orf12*	*DCX*	*EXOSC8*	*GFAP*	*IKBKAP*	*LYST*	*NTRK1*	*POLR3B*	*RTN2*	*SLC52A2*	*TBR1*	*UBA1*
AKT1	*ATL3*	*CLPP*	*DYNC1H1*	*FKRP*	*HINT1*	*KIF1C*	*MR1*	*PEX10*	*PRPS1*	*SETX*	*SPG7*	*TRIM2*	*VRK1*
AKT3	*ATM*	*COASY*	*EDN3*	*FKTN*	*HK1*	*KIF21A*	*MRE11A*	*PFN1*	*PRRT2*	*SGCE*	*SPR*	*TRIP4*	*WDR45*
ALDH18A1	*ATP1A3*	*COL4A1*	*EGR2*	*FLNA*	*HNRNPA1*	*KIF2A*	*MTMR2*	*PGAP1*	*PRX*	*SH3TC2*	*SPTBN2*	*TRPV4*	*WDR48*
ALDH3A2	*ATP2B3*	*COL4A2*	*EIF2B1*	*FLRT1*	*HNRNPA2B1*	*KIF5A*	*MTPAP*	*PIK3R5*	*PSAP*	*SIGMAR1*	*SPTLC1*	*TSEN2*	*WDR62*
ALS2	*ATP2B4*	*COX6A1*	*EIF2B2*	*FLVCR1*	*HNRNPUL1*	*KIF5C*	*MYH14*	*PLA2G6*	*PTEN*	*SIL1*	*SPTLC2*	*TSEN34*	*WNK1*
AMPD2	*ATP6AP2*	*CPT1C*	*EIF2B3*	*FTL*	*HNRNPUL2*	*KLC2*	*MYH7*	*PLEKHG5*	*RAB3GAP2*	*SLC12A6*	*SPTLC3*	*TSEN54*	*WWOX*
ANG	*ATP7A*	*CSF1R*	*EIF2B4*	*FUS*	*HOXD10*	*KLC4*	*NAGLU*	*PLOD1*	*RAB7A*	*SLC16A2*	*SQSTM1*	*TTBK2*	*YARS*
ANO10	*ATR*	*CTDP1*	*EIF2B5*	*FXN*	*HPCA*	*KTN1*	*NDE1*	*PLP1*	*REEP1*	*SLC17A5*	*STUB1*	*TTPA*	*YWHAE*
ANO3	*B3GALNT2*	*CUL3*	*ELOVL4*	*GAD1*	*HSPB1*	*L1CAM*	*NDRG1*	*PMP2*	*REEP2*	*SLC18A3*	*SYNE1*	*TTR*	*ZBTB18*
AP4B1	*B4GALNT1*	*CWF19L1*	*ELOVL5*	*GALC*	*HSPB3*	*LARGE*	*NEFL*	*PMP22*	*RELN*	*SLC1A3*	*SYT14*	*TUBA1A*	*ZFR*
AP4E1	*BCAP31*	*CYP2U1*	*ENTPD1*	*GAN*	*HSPB8*	*LAS1L*	*NGF*	*PMPCA*	*RNASEH2B*	*SLC20A2*	*TAF1*	*TUBB*	*ZFYVE26*
AP4M1	*BEAN1*	*CYP7B1*	*EOMES*	*GARS*	*HSPD1*	*LITAF*	*NIPA1*	*PNKP*	*RNASET2*	*SLC25A46*	*TARDBP*	*TUBB2B*	*ZFYVE27*
AP4S1	*BICD2*	*DARS2*	*ERLIN1*	*GBA2*	*IBA57*	*LMNA*	*NOP56*	*PNPLA6*	*RNF170*	*SLC2A1*	*TBCD*	*TUBB3*	
AP5Z1	*BSCL2*	*DCAF8*	*ERLIN2*	*GCH1*	*IFIH1*	*LMNB1*	*NOTCH3*	*POLG*	*RNF216*	*SLC33A1*	*TBK1*	*TUBB4A*	
APTX	*C12orf65*	*DCTN1*	*EXOSC3*	*GDAP1*	*IGHMBP2*	*LRSAM1*	*NT5C2*	*POLR3A*	*RPIA*	*SLC52A1*	*TBP*	*TUBG1*	

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