Becker and Duchenne Muscular Dystrophy Test

The Blueprint Genetics Becker and Duchenne Muscular Dystrophy test is a sequencing analysis of *DMD* gene aiming to identify disease-causing mutations that are missed in the classical deletion-duplication analyses.

The Becker and Duchenne Muscular Dystrophy Test provides a high quality read-out of all exons of *DMD* gene. Our OS-Seq™ technology provides high coverage clinical grade sequencing and enables reliable diagnostics for patients with significantly lower costs and faster turnaround time (basic service TAT 21 days and Express service TAT 7-10 days). The Becker and Duchenne Muscular Dystrophy Test has undergone rigorous validation process during its evolution at Blueprint Genetics. Our unique sequencing technology combined with in-house built bioinformatics pipeline with Becker and Duchenne muscular dystrophy mutation and knowledge database, together with our experienced team of geneticists and clinicians, enables efficient diagnostics for Becker and Duchenne muscular dystrophy patients. Our variant classification schemes and clinical interpretation processes have been developed and validated with thousands of patients with hereditary cardiovascular disease. Blueprint Genetics publically shares all classified variants to improve future diagnostics (ClinVar; http://www.ncbi.nlm.nih.gov/clinvar/). Our mission is to improve the quality of diagnostics and management of Becker and Duchenne muscular dystrophy patients and their families.

Genes Covered by Panel

The test covers the sequencing of *DMD* gene. All protein coding exons and exon-intron boundaries are covered. Sequencing is also targeted to other regions with reported pathogenic or likely pathogenic mutations.

Description of Test

Blueprint genetics offers the Becker and Duchenne Muscular Dystrophy Test that covers the sequencing of *DMD* gene.

Coverage

In our latest validation of the Becker and Duchenne Muscular Dystrophy Test, median sequencing depth in the target region was 295x on a nucleotide level and 99.40% of the nucleotides had at least 15x coverage.

Analytical validity

Analytical validation is a continuous process at Blueprint Genetics. Our mission is to improve the quality of the sequencing process and our standardized validation process follows each modification. In our latest validation the Becker and Duchenne Muscular Dystrophy Test had sensitivity of 0.990 and specificity of 1.000 to detect single nucleotide polymorphisms. Our panel had also good performance to detect insertions and deletions. Sensitivity was 1.000 for both indels ranged 1-5 bp and 6-19 bp. This panel has not been validated to identify larger deletions, insertions or complex rearrangements.

Diagnostic yield

Blueprint Genetics provides genetic diagnostics for hundreds of hospitals and clinics around the world. The diagnostic yield varies substantially between hospitals and countries. Diagnostic yield with Becker and Duchenne Muscular Dystrophy Test cannot be reliably estimated as the number of patients analyzed, to date, is not high enough. Please review our variant interpretation strategy here.
Bioinformatics

We have developed a unique Becker and Duchenne muscular dystrophy sequence analyzer and interpretation pipeline. In the core of our bioinformatics lies our in-house created and curated Becker and Duchenne muscular dystrophy mutation database, which is a synthesis of original publications on Becker and Duchenne muscular dystrophy and existing mutation databases. The analysis pipeline includes rigorous quality control steps to ensure validity and consistency of results, and incorporates gene variability data from thousands of publicly available human reference sequences in order to eliminate false positive findings and deliver data of the highest relevance. Reference databases currently used are 1000 Genomes (http://www.1000genomes.org), NHLBI GO Exome Sequencing Project (ESP; http://evs.gs.washington.edu/EVS), Exome Aggregation Consortium (ExAC; http://exac.broadinstitute.org). In addition, we apply the following in silico variant prediction tools in our analyses: SIFT (http://sift.jcvi.org), Polyphen (http://genetics.bwh.harvard.edu/pph2/), and Mutation Taster (http://www.mutationtaster.org).

Through our online ordering and statement system, Nucleus, the customer can access specific details of the analysis of the patient. This includes coverage and quality specifications and other information on the analysis. This represents our mission to build fully transparent diagnostics where customer has easy access to the details of the analysis.

Clinical Interpretation

In addition to our cutting-edge proprietary sequencing technology and bioinformatics pipeline, BpG also provides the customers with the best-informed clinical statement on the market. Clinical interpretation requires fundamental clinical and genetic understanding on hereditary cardiovascular diseases. At Blueprint Genetics our geneticists and cardiologists, who together, evaluate the results from the sequence analysis pipeline in the context of phenotype information provided in the requisition form, prepare the clinical statement. Our goal is to provide clinically meaningful statements that are understandable for all medical professionals, even without training in genetics.

Variants reported in the statement are always classified using the Blueprint Genetics Classification Scheme modified from the ACMG guidelines (Richards et al. 2015), which has been developed by evaluating existing literature, databases and with thousands of clinical cases analyzed in our laboratory. Variant classification forms the corner stone of clinical interpretation and following patient management decisions. Our statement also provides allele frequencies in existing databases and in silico predictions. We also provide PubMed IDs to the articles or submission numbers to public databases that have been used in the interpretation of the detected variants. In our conclusion, we summarize all the existing information and provide our rationale for the classification of the variant.

A final component of the analysis is the Sanger confirmation of clinically relevant variants, variants classified as likely pathogenic or pathogenic. This does not only bring confidence to the results obtained by our NGS solution but establishes the mutation specific test for family members. Sanger sequencing is also used occasionally with other variants reported in the statement. In the case of variant of unknown significance (VUS) we do not recommend risk stratification based on the genetic finding. Furthermore, in the case VUS we do not recommend use of genetic information in patient management or genetic counseling. For some cases Blueprint Genetics offers a special service to investigate the role of identified VUS.

We constantly follow the development on the field of Becker and Duchenne muscular dystrophy and adapt new relevant information and findings to our diagnostics. Relevant novel discoveries can be rapidly translated and adopted into our diagnostics without delay. These processes ensure that our diagnostic panels and clinical statements remain the most up-to-date and relevant on the market.

Sample Requirements

- 3ml of EDTA blood
- Purified DNA 10μg
- Saliva (Oragene DNA OG-500 or OGD-500 kit, DNA Genotek)

Details for sample preparations and sending are found here.

About the Disorder

Muscular dystrophies (MDs) are genetic conditions characterized by progressive muscle weakness and wasting. The Duchenne (DMD) and Becker MD (BMD) are related conditions caused by different mutations in the same defective gene, namely dystrophin (DMD) and thus called dystrophinopathies. These forms of muscular dystrophy occur almost exclusively in
males. DMD and BMD have similar signs and symptoms but differences in their severity, age of onset, and rate of progression. Affected children may have delayed gain of age related motor skills such as sitting, standing, and walking. They are usually wheelchair-dependent by adolescence. Clinical presentation of Becker muscular dystrophy is usually milder and more varied. Muscle weakness tends to become apparent later in childhood or in adolescence and worsens at a much slower rate. Both the Duchenne and Becker MDs are associated with a dilated cardiomyopathy, which typically begins in adolescence and presents with arrhythmias, poor exercise tolerance, and heart failure. These heart problems progress rapidly and become life-threatening in many cases. Life expectancy in males with Duchenne or Becker muscular dystrophy is typically 20 and 40 years. Up to 8% of female ‘carriers’ with Duchenne type mutations have isolated dilated cardiomyopathy without skeletal muscle involvement.

Existing therapies for these muscular dystrophies are limited to corticosteroids for membrane stabilization and mediating anti-inflammatory effects and those targeting exclusively to DMD cardiomyopathy such as ACE inhibitors and betablockers. Moreover, influenza and pneumococcal vaccination, nutritional aspects, physiotherapy and D-vitamin are used to prevent secondary complications. Interestingly, there is new hope from a recent discovery at phase 1-2a trials showing some benefit from antisense oligonucleotide treatment to turn Duchenne phenotype to less aggressive Becker phenotype. In this therapy antisense oligonucleotide, as structural analogs of DNA, are used to skip the defective exon of the DMD gene (e.g. (PRO051: exon-51 skip therapy), which leads to expression of at least partially functional truncated protein. An early phase 1-2a study showed improved results in the 6-minute walk test (Goemans et al. 2011). These of experimental therapies require definite molecular genetics diagnosis to identify the patients with mutations suitable for the targeted therapy.

For males with clinical findings of DMD or BMD and an elevated serum CK concentration, molecular genetic testing as deletion/duplication analysis should be initiated. If a large gene defect is not identified, sequence analysis is indicated. However, differentiation between the two clinical conditions may be difficult even if a disease-causing DMD mutation is identified. For example, deletions in exons 3-7 are common in both phenotypes (Aartsma-Rus et al 2006b). A reading frame ‘rule’ can be often employed in differential diagnostics. Mutations that do not alter the reading frame (in-frame deletions/duplications) generally associate with the BMD, and those altering the reading frame (out-of-frame) with DMD phenotype (Monaco et al 1988). Exceptions to the “reading frame rule” have been documented to occur more at BMD compared to DMD and these have been discussed in detail in recent articles (Aartsma-Rus et al. 2006b, Kesari et al. 2008, Takeshima et al. 2010). If no disease-causing DMD mutation is identified, skeletal muscle biopsy should be considered for Western blot and immunohistochemistry studies of dystrophin. Diagnostic scheme for boys with DMD and other X-linked disorders and for girls with classic DMD phenotype is different (Darras et al. 2011). Carrier testing for at-risk female relatives should be performed by utilization of same test that identified the index patient’s mutation.

References


