

Blue native polyacrylamide gel electrophoresis for mitochondrial myopathies

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Next review date: 10/2025

Policy contains: Blue native polyacrylamide gel electrophoresis, mitochondrial myopathy.

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Coverage policy

Blue native polyacrylamide gel electrophoresis for the diagnosis of mitochondrial myopathies is considered experimental/investigational and not clinically proven.

Limitations

No limitations were identified during the writing of this policy.

Alternative covered services

- Muscle biopsies.
- Electromyography.
- Nerve conduction studies.

Background

Mitochondria are multifunctional organelles at a cellular level that plays an important role in energy production, cellular metabolism regulation, signalling, apoptosis (cellular death), and aging. The energy production is dependent on an oxidative phosphorylation process that combines respiration with ATP synthesis in humans and are vital for survival (Konovalova, 2019). Mitochondrial disease exists in 1 of 4,300 Americans, affecting all age groups, and can be difficult to diagnose (Children's Hospital of Philadelphia, 2022).

Mitochondrial myopathies are muscular problems of varying severity marked by muscle fatigue, weakness, and exercise intolerance. The disorder may include weakness and wasting in face and neck muscles, swallowing difficulty, slurred speech, and arm/leg weakness. Myopathies are genetic disorders, in which ATP-deprived cells accumulate unused fuel molecules and destructive free radical/reactive oxygen. Fuel molecules can generate harmful byproducts such as lactic acid, which can create muscle fatigue and damage muscle and nerve tissue.

Syndromes associated with mitochondrial disease can include:

Barth syndrome.

Chronic progressive external ophthalmoplegia.

Kearns-Sayre syndrome.

Maternally inherited Leigh syndrome.

Mitochondrial DNA depletion syndromes.

Mitochondrial encephalomyopathy, lactic acidosis, and stroke-like episodes.

Mitochondrial neurogastrointestinal encephalomyopathy.

Myoclonus epilepsy with ragged red fibers.

Neuropathy, ataxia, and retinitis pigmentosa.

Pearson syndrome (National Institute of Neurological Disorders and Stroke, no date given).

Diagnosis in symptomatic patients includes:

- Physical exam, including strength and endurance tests.
- Medical history, including a family history to assess any genetic links to the disorder.
- Neurological exam, including reflexes, vision, speech, and cognitive skills.
- Lab tests for diabetes, liver, and kidney disorders, plus an electrocardiogram.
- Imaging of organs, bones, and tissues.
- Lactic acid testing for lactic acidosis.
- Muscle biopsy, to identify any cells with excessive mitochondria.
- Genetic testing for mutations known to cause mitochondrial myopathy (National Institute of Neurological Disorders and Stroke, no date given).

Treatments of mitochondrial myopathy attempt to fix or bypass defective mitochondria through nutritional supplements that aid in ATP production. These supplements include creatine, carnitine, and coenzyme Q10.

Blue native polyacrylamide gel electrophoresis is a method of complexome profiling that separates multi-protein complexes to determine size, composition, and relative abundance of these complexes. The main advantage to using this is that relatively low amounts of biological material are needed; and the high resolution separation of the native proteins (Cabrera-Orefice, 2022). The test separates proteins according to their hydrodynamic size and shape in a polyacrylamide matrix (Fiala, 2011). It requires no specialized equipment, is compatible with most protein detection methods, and has been used since 1991 (Yeh, 2010).

Potential applications of blue native electrophoresis include immunological and receptor studies, biogenesis and assembly of membrane protein complexes, protein import into organelles, dynamics of proteasomes, proteome and subproteome investigations, and identification and quantification of mitochondrial alterations in apoptosis, carcinogenesis, and neurodegenerative disorders like Parkinson's disease, Alzheimer's disease, and a variety of mitochondrial encephalomyopathies (Wittig, 2008).

One study states that blue native polycrylamide gel electrophoresis "has been used in many studies to assess the oligomeric state of mitochondrial carriers" due to its low cost and small size (Crichton, 2013).

Findings

No professional medical society guidelines addressing indications for polyacrylamide gel electrophoresis for mitochondrial myopathies exist, nor are any systematic reviews/meta-analyses available in the medical literature.

A review of 30 cases of child hypertrophic cardiomyopathy caused by mitochondrial disease employed blue native polyacrylamide gel electrophoresis, plus other biological and genetic techniques. Authors state that “establishing a genetic diagnosis in mitochondrial cardiomyopathy is challenging and achieved in only a minority of cases because of complex genetics,” raising the question of efficacy in practice (Fassone, 2011).

A study of symptomatic patients (n = 390, 115 of whom were determined to have mitochondrial disease), found the ability of blue native polyacrylamide gel electrophoresis to distinguish mitochondrial and non-mitochondrial cases was statistically significant, as a step prior to genetic testing (Kerr, 2020).

A review analyzed mitochondrial function in tissues from 45 liver transplantation patients using blue native polyacrylamide gel electrophoresis with immunodetection of respiratory chain complexes I-V, biochemical activity of respiratory chain complexes II and IV and quantification of mitochondrial DNA. Ten of 40 patients (25%) had a defect of one or more respiratory chain enzyme complexes; 20 patients (44%) had low activity of complex II and/or IV; and ten (22%) had a reduced mtDNA copy number (Lane, 2016).

A study analyzed 11 fibroblast cell lines from patients with inherited complex I deficiency, which accounts for over 30% of mitochondrial diseases. In patient cells carrying a mutation located in the matrix arm of complex I deficiency, blue native polyacrylamide gel electrophoresis found a significant reduction of fully assembled complex I enzyme, which authors said showed “suitability” in evaluating the deficiency (Leman, 2015).

A study identified 675 Japanese patients with profound lactic acidemia, or with symptoms of multiple-organ origin without lactic acidemia. Tests to identify these patients were respiratory chain enzyme activity assay and blue native polyacrylamide gel electrophoresis. Polymerase chain reaction diagnosed 232 of these patients with probable or definite mitochondrial respiratory chain disorders (Yamazaki, 2014).

A review of four studies concluded that blue native polyacrylamide gel electrophoresis was suitable for identifying multi-protein complexes and determining their size, composition, and relative abundance (Fiala, 2011).

A study of 50 adults with multiple mitochondrial DNA deletions in skeletal muscle analyzed ribonucleotide reductase using western blot and blue native polyacrylamide gel electrophoresis. Four percent of this group harbored RRM2B mutations caused by ribonucleotide reductase dysfunction, and the authors suggested screening to diagnose adults with multiple mitochondrial deletions (Pithceathly, 2011).

A study of 66 tissue samples in 53 patients identified catalytically active subcomplexes of complex V, site of the final step in oxidative phosphorylation, using blue native polyacrylamide gel electrophoresis followed by activity staining in the gel. In 55% (29 of 53) patients, a mitochondrial DNA defect was identified (Smet, 2009).

A number of articles in the medical literature address use of blue native polyacrylamide gel electrophoresis in animals (often rats) and plants (such as potatoes or rice), or explore its efficacy in human tissues using sample sizes of less than 10 patients, often single case studies, which fail to document the technique’s use in standard clinical practice.

An article describes blue native polyacrylamide gel electrophoresis as “proven to be particularly invaluable... providing researchers with a facile approach for analyzing the assembly, total abundance, and residual enzymatic activity of individual OXPHOS (oxidative phosphorylation) complexes,” suggesting its primary use is research, not practice (Leary, 2012).

In 2024, no new relevant articles were found.

References

On May 11, 2024, we searched PubMed and the databases of the Cochrane Library, the U.K. National Health Services Centre for Reviews and Dissemination, the Agency for Healthcare Research and Quality, and the Centers for Medicare & Medicaid Services. Search terms were “blue native polyacrylamide gel electrophoresis,” and “mitochondrial myopathy.” We included the best available evidence according to established evidence hierarchies (typically systematic reviews, meta-analyses, and full economic analyses, where available) and professional guidelines based on such evidence and clinical expertise.

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Policy updates

6/2021: initial review date and clinical policy effective date: 7/2021

6/2022: Policy references updated.

6/2023: Policy references reviewed – no current updates.

6/2024: Policy references reviewed – no current updates warranted.