

Peroxisomal Disorders Panel

Panel Gene List: *ABCD1, ACOX1, AGPS, AGXT, AMACR, DNM1L, GNPAT, HSD17B4, PEX1, PEX2, PEX3, PEX5, PEX6, PEX7, PEX10, PEX11B, PEX12, PEX13, PEX14, PEX16, PEX19, PEX26, PHYH, SCP2, TRIM37*

Peroxisomal disorders overview:

Peroxisomal disorders comprise two heterogeneous subgroups of conditions: peroxisomal biogenesis disorders (PBD) which are characterized by defects in peroxisome synthesis, assembly and biochemical functions, and disorders of single peroxisomal enzymes and beta-oxidation deficiencies which are involved in ether lipid biosynthesis, phytanic, pristanic, and pipercolic acid catabolism, fatty acid beta-oxidation and other functions localized to peroxisomes.

Clinical features of peroxisomal biogenesis disorders:

Peroxisomal biogenesis disorders consist of i) Zellweger spectrum disorders, including Zellweger syndrome, neonatal adrenoleukodystrophy, and infantile Refsum syndrome, and ii) rhizomelic chondrodysplasia punctata. Zellweger spectrum disorder involves a range of symptoms such as hypotonia, liver dysfunction, hearing loss, retinal dystrophy, optic nerve abnormalities, seizures, leukodystrophy, chondrodysplasia punctata, and characteristic facial features. While children with Zellweger syndrome are typically diagnosed in the neonatal period, individuals with neonatal adrenoleukodystrophy and Refsum syndrome may present with less severe symptoms and may be diagnosed later in childhood. Rhizomelic chondrodysplasia punctata is characterized by rhizomelia, epiphyseal and metaphyseal abnormalities, intellectual disability, seizures, cataracts, coronal clefts, contractures, characteristic facial features, and pulmonary hypoplasia.^{1,2}

Clinical features of peroxisomal enzymes and beta-oxidation deficiencies:

Disorders of peroxisomal single enzyme deficiencies include adult Refsum syndrome, X-linked adrenoleukodystrophy, rhizomelic chondrodysplasia punctata type 2 and type 3, acyl-CoA oxidase deficiency, D-bifunctional enzyme deficiency, alpha-methylacyl-CoA racemase deficiency, *DNM1L*-related leukoencephalopathy, and mulibrey nanism. The clinical features of peroxisomal enzyme deficiencies vary depending on the specific disorder, however many patients may present with neurological deficits and symptoms similar to Zellweger spectrum disorder. A diagnosis of peroxisomal disorders is based on clinical features, biochemical studies that include analysis of very long chain fatty acids, phytanic acid, pipercolic acid, pristanic acid, plasmalogens and bile acids, and complementation analysis in fibroblasts.³

Utility of molecular testing:

The diagnosis of peroxisomal disorders is challenging; many PBDs have overlapping clinical and biochemical features which, in the absence of complementation analysis in fibroblasts, may make it difficult to identify the underlying molecular etiology. In addition, biochemical analysis may not always reveal evidence for abnormal peroxisome function.^{1,4,5} Due to the biochemical complexity, genetic heterogeneity, and the need for a fibroblast biopsy for complementation studies, many clinicians are using peroxisomal-targeted Next generation sequencing panels in the diagnosis of patients. In addition, clarification of the underlying genotype may have prognostic value, and is essential for genetic counseling and subsequent family studies.

Genetics:

With the exception of X-linked adrenoleukodystrophy and *DNM1L*-associated leukoencephalopathy, peroxisomal disorders are inherited in an autosomal recessive manner. X-linked adrenoleukodystrophy is caused by pathogenic variants in the *ABCD1* gene, while *DNM1L*-associated leukoencephalopathy has been reported with both autosomal dominant and autosomal recessive inheritance.

Test Methods:

Using genomic DNA from the submitted specimen, the complete coding regions and splice site junctions of the genes on this panel are enriched using a proprietary targeted capture system developed by GeneDx for next-generation sequencing with CNV calling (NGS-CNV). The enriched targets are simultaneously sequenced with paired-end reads on an Illumina platform. Bi-directional sequence reads are assembled and aligned to reference sequences based on NCBI RefSeq transcripts and human genome build GRCh37/UCSC hg19. After gene specific filtering, data are analyzed to identify sequence variants and most deletions and duplications involving coding exons. Alternative sequencing or copy number detection methods are used to analyze regions with inadequate sequence or copy number data. Concurrently, multiplex ligation-dependent probe amplification (MLPA) is performed to evaluate for an exon-level deletion or duplication of the *ABCD1* gene. This test does not include deletion/duplication testing of exon 9 of *ABCD1*. Reportable variants include pathogenic variants, likely pathogenic variants and variants of uncertain significance. Likely benign and benign variants, if present, are not routinely reported but are available upon request.

The technical sensitivity of sequencing is estimated to be > 99% at detecting single nucleotide events. It will not reliably detect deletions greater than 20 base pairs, insertions or rearrangements greater than 10 base pairs, or low-level mosaicism. This test may not reliably detect copy number variants of less than 500 base pairs or low level mosaicism and cannot identify balanced chromosome aberrations. Assessment of exon-level copy number events is

dependent on the inherent sequence properties of the targeted regions, including shared homology and exon size.

Clinical Test Sensitivity:

The clinical sensitivity of sequencing and deletion/duplication analysis of the genes included in this panel depends in part on the patient's clinical phenotype. For example, sequence analysis of 14 peroxin genes associated with PBDs is estimated to detect ~96% of affected individuals; ~70% of cases are caused by pathogenic variants in *PEX1*, while ~26% of cases are due to pathogenic variants in *PEX6*, *PEX10*, *PEX12*, or *PEX26*.^{1,2,3} In a large study of patients with a clinical diagnosis of X-ALD, sequencing analysis of *ABCD1* detected pathogenic variants in 95% of individuals.⁶ Deletions have been detected in approximately 3% of individuals with X-ALD.⁷

Class of Disorders	Gene Name	Associated Disorder(s)	OMIM #
Peroxisomal biogenesis disorders	<i>PEX1</i>	Zellweger spectrum disorder Heimler syndrome 1	602136
	<i>PEX2</i>	Zellweger spectrum disorder	614866
	<i>PEX3</i>	Zellweger spectrum disorder	617370
	<i>PEX5</i>	Zellweger spectrum disorder Rhizomelic chondrodysplasia punctata type 5	600414
	<i>PEX6</i>	Zellweger spectrum disorder Heimler syndrome 2	601498
	<i>PEX7</i>	Rhizomelic chondrodysplasia punctata type 1/ Refusm syndrome	614879
	<i>PEX10</i>	Zellweger spectrum disorder	614870
	<i>PEX11B</i>	Zellweger spectrum disorder	614920
	<i>PEX12</i>	Zellweger spectrum disorder	614859
	<i>PEX13</i>	Zellweger spectrum disorder	614883
	<i>PEX14</i>	Zellweger spectrum disorder	614887
	<i>PEX16</i>	Zellweger spectrum disorder	614876
	<i>PEX19</i>	Zellweger spectrum disorder	614886
	<i>PEX26</i>	Zellweger spectrum disorder	614872
Disorders of single peroxisomal enzymes and beta-oxidation deficiencies	<i>ABCD1</i>	X-linked adrenoleukodystrophy	300100
	<i>ACOX1</i>	Peroxisomal acyl-CoA oxidase deficiency	264470
	<i>AGPS</i>	Rhizomelic chondrodysplasia punctata type 3	600121
	<i>AGXT</i>	Primary hyperoxaluria	259900
	<i>AMACR</i>	Alpha-methylacyl-CoA racemase deficiency	614307
	<i>DNM1L</i>	<i>DNM1L</i> -associated leukoencephalopathy	614388
	<i>GNPAT</i>	Rhizomelic chondrodysplasia punctata type 2	222765

	<i>HSD17B4</i>	D-bifunctional protein deficiency Perrault syndrome 1	601860
	<i>PHYH</i>	Adult Refsum syndrome	266500
	<i>SCP2</i>	Sterol carrier protein deficiency	604105
	<i>TRIM37</i>	Mulibrey nanism	253250

References:

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