Blueprint Genetics

Purine and Pyrimidine Metabolism Disorders Panel

Test code: ME2401

Is ideal for patients with a clinical suspicion of an inborn error in the metabolism of purines or pyrimidines. The genes on this panel are included in the Comprehensive Metabolism Panel.

About Purine and Pyrimidine Metabolism Disorders

Genetic disorders of purine and pyrimidine metabolism are under-reported and infrequently mentioned in the literature of other inborn errors of metabolism. These disorders represent a broad spectrum of clinical manifestations and challenging diagnostic problems. They may affect any system in a variety of ways, and often mimic other, more recognizable disorders. Owing to these factors and limited awareness, these disorders may often be misdiagnosed or remain undiagnosed. Furthermore, biochemical analysis of these disorders requires special techniques that are not widely available. More than 30 defects in human purine and pyrimidine metabolic pathways have been identified thus far. Many are benign, but about half are associated with severe clinical manifestations, some causing major morbidity and mortality. The most common of purine and pyrimidine metabolism disorders is the deficiency of hypoxanthine phosphoribosyl transferase that also causes hyperuricaemia in childhood.

Availability

Results in 3-4 weeks

Gene set description

Genes in the Purine and Pyrimidine Metabolism Disorders Panel and their clinical significance

Gene	Associated phenotypes	Inheritance	ClinVar	HGMD
ADA	Severe combined immunodeficiency due to adenosine deaminase deficiency	AR	49	93
ADSL	Adenylosuccinase deficiency	AR	24	57
AMPD1	Myoadenylate deaminase deficiency	AR	5	10
APRT	Adenine phosphoribosyltransferase deficiency	AR	11	47
ATIC	AICAR transformylase/IMP cyclohydrolase deficiency	AR	2	6
DHODH	Postaxial acrofacial dysostosis (Miller syndrome)	AR	8	20
DPYD	5-fluorouracil toxicity, Developmental delay with or without dysmorphic facies and autism	AD/AR	62	86
DPYS	Dihydropyriminidase deficiency	AR	8	29
GPHN	Hyperekplexia, Molybdenum cofactor deficiency	AD/AR	35	20
HPRT1	Lesch-Nyhan syndrome, Kelley-Seegmiller syndrome	XL	72	427
MOCOS	Xanthinuria, type II	AR	3	5
MOCS1*	Molybdenum cofactor deficiency	AR	7	35
NT5C3A	Uridine 5-prime monophosphate hydrolase deficiency, hemolytic anemia due to	AR	10	28

PNP	Purine nucleoside phosphorylase deficiency	AR	11	33
PRPS1*	Phosphoribosylpyrophosphate synthetase I superactivity, Arts syndrome, Charcot-Marie-Tooth disease, X-linked recessive, 5, Deafness, X-linked 1	XL	27	32
REN	Hyperuricemic nephropathy, Hyperproreninemia, familial, Renal tubular dysgenesis	AD/AR	9	18
TPMT*	Thiopurine S-methyltransferase deficiency	AR		16
UMOD	Familial juvenile hyperuricemic nephropathy, Glomerulocystic kidney disease with hyperuricemia and isosthenuria, Medullary cystic kidney disease 2	AD	33	108
UMPS	Orotic aciduria	AR	3	12
UPB1	Beta-ureidopropionase deficiency	AR	5	17
XDH	Xanthinuria, type I	AR	10	21

*Some regions of the gene are duplicated in the genome. Read more.

The gene has suboptimal coverage (means <90% of the gene's target nucleotides are covered at >20x with mapping quality score (MQ>20) reads), and/or the gene has exons listed under Test limitations section that are not included in the panel as they are not sufficiently covered with high quality sequence reads.

The sensitivity to detect variants may be limited in genes marked with an asterisk (*) or number sign (#)

Gene refers to the HGNC approved gene symbol; Inheritance refers to inheritance patterns such as autosomal dominant (AD), autosomal recessive (AR), mitochondrial (mi), X-linked (XL), X-linked dominant (XLD) and X-linked recessive (XLR); ClinVar refers to the number of variants in the gene classified as pathogenic or likely pathogenic in this database (ClinVar); HGMD refers to the number of variants with possible disease association in the gene listed in Human Gene Mutation Database (HGMD). The list of associated, gene specific phenotypes are generated from CGD or Mitomap databases.

Non-coding disease causing variants covered by the panel

Gene	Genomic location HG19	HGVS	RefSeq	RS-number
ADA	Chr20:43248503	c.1079-15T>A	NM_000022.2	rs387906268
ADA	Chr20:43249076	c.976-34G>A	NM_000022.2	
ADSL	Chr22:40742514	c.-49T>C	NM_000026.2	
HPRT1	ChrX:133594415	c.27+47C>T	NM_000194.2	
HPRT1	ChrX:133625464	c.402+1229A>G	NM_000194.2	
HPRT1	ChrX:133628822	c.485+1202T>A	NM_000194.2	
HPRT1	ChrX:133632625	c.533-13T>G	NM_000194.2	
MOCS1	Chr6:39874534	c.*365_*366delAG	NM_005943.5	rs397518419
MOCS1	Chr6:39876810	c.*7+6T>C	NM_005943.5	
MOCS1	Chr6:39894006	c.251-418delT	NM_005943.5	
PNP	Chr14:20942914	c.286-18G>A	NM_000270.3	

REN	Chr1:204129817	c.374-12_374-11delTCinsAG	NM_000537.3

Test Strengths

The strengths of this test include:

- CAP accredited laboratory
- CLIA-certified personnel performing clinical testing in a CLIA-certified laboratory
- Powerful sequencing technologies, advanced target enrichment methods and precision bioinformatics pipelines ensure superior analytical performance
- Careful construction of clinically effective and scientifically justified gene panels
- Some of the panels include the whole mitochondrial genome (please see the Panel Content section)
- Our Nucleus online portal providing transparent and easy access to quality and performance data at the patient level
- Our publicly available analytic validation demonstrating complete details of test performance
- ~2,000 non-coding disease causing variants in our clinical grade NGS assay for panels (please see 'Non-coding disease causing variants covered by this panel' in the Panel Content section)
- Our rigorous variant classification scheme
- Our systematic clinical interpretation workflow using proprietary software enabling accurate and traceable processing of NGS data
- Our comprehensive clinical statements

Test Limitations

Genes with partial, or whole gene, segmental duplications in the human genome are marked with an asterisk (*) if they overlap with the UCSC pseudogene regions. The technology may have limited sensitivity to detect variants in genes marked with these symbols (please see the Panel content table above).

This test does not detect the following:

- Complex inversions
- Gene conversions
- Balanced translocations
- Some of the panels include the whole mitochondrial genome but not all (please see the Panel Content section)
- Repeat expansion disorders unless specifically mentioned
- Non-coding variants deeper than ±20 base pairs from exon-intron boundary unless otherwise indicated (please see above Panel Content / non-coding variants covered by the panel).

This test may not reliably detect the following:

- Low level mosaicism in nuclear genes (variant with a minor allele fraction of 14.6% is detected with 90% probability)
- Stretches of mononucleotide repeats
- Low level heteroplasmy in mtDNA (>90% are detected at 5% level)
- Indels larger than 50bp
- Single exon deletions or duplications
- Variants within pseudogene regions/duplicated segments
- Some disease causing variants present in mtDNA are not detectable from blood, thus post-mitotic tissue such as skeletal muscle may be required for establishing molecular diagnosis.

The sensitivity of this test may be reduced if DNA is extracted by a laboratory other than Blueprint Genetics.

For additional information, please refer to the Test performance section and see our Analytic Validation.

Test performance

The Blueprint Genetics purine and pyrimidine metabolism disorders panel covers classical genes associated with Lesch-Nyhan syndrome, purine nucleoside phosphorylase deficiency, severe combined immunodeficiency due to adenosine deaminase deficiency, adenylosuccinase deficiency, adenine phosphoribosyltransferase deficiency, beta-ureidopropionase deficiency and uridine 5-prime monophosphate hydrolase deficiency, hemolytic anemia due to. The genes on the panel have been carefully selected based on scientific literature, mutation databases and our experience.

Our panels are sliced from our high-quality whole exome sequencing data. Please see our sequencing and detection performance table for different types of alterations at the whole exome level (Table).

Assays have been validated for different starting materials including EDTA-blood, isolated DNA (no FFPE), saliva and dry blood spots (filter card) and all provide high-quality results. The diagnostic yield varies substantially depending on the assay used, referring healthcare professional, hospital and country. Blueprint Genetics' Plus Analysis (Seq+Del/Dup) maximizes the chance to find a molecular genetic diagnosis for your patient although Sequence Analysis or Del/Dup Analysis may be a cost-effective first line test if your patient's phenotype is suggestive of a specific mutation type.

The genes on the panel have been carefully selected based on scientific literature, mutation databases and our experience.

Our panels are sectioned from our high-quality, clinical grade NGS assay. Please see our sequencing and detection performance table for details regarding our ability to detect different types of alterations (Table).

	Sensitivity % (TP/(TP+FN)	Specificity %
Single nucleotide variants	99.89% (99,153/99,266)	>99.9999%
Insertions, deletions and indels by sequence analysis		
1-10 bps	96.9% (7,563/7,806)	>99.9999%
11-50 bps	99.13% (2,524/2,546)	>99.9999%
Copy number variants (exon level dels/dups)		
1 exon level deletion (heterozygous)	100% (20/20)	NA
1 exon level deletion (homozygous)	100% (5/5)	NA
1 exon level deletion (het or homo)	100% (25/25)	NA
2-7 exon level deletion (het or homo)	100% (44/44)	NA
1-9 exon level duplication (het or homo)	75% (6/8)	NA
Simulated CNV detection		
5 exons level deletion/duplication	98.7%	100.00%
Microdeletion/-duplication sdrs (large CNVs, n=37))		
Size range (0.1-47 Mb)	100% (37/37)	

The performance presented above reached by Blueprint Genetics high-quality, clinical grade NGS sequencing assay with the following coverage metrics

Mean sequencing depth	143X

Nucleotides with >20x sequencing coverage (%)	99.86%

Performance of Blueprint Genetics Mitochondrial Sequencing Assay.

	Sensitivity % (TP/(TP+FN)	Specificity
ANALYTIC VALIDATION (NA samples; n=4)		
Single nucleotide variants		
Heteroplasmic (45-100%)	100.0% (50/50)	100.0%
Heteroplasmic (35-45%)	100.0% (87/87)	100.0%
Heteroplasmic (25-35%)	100.0% (73/73)	100.0%
Heteroplasmic (15-25%)	100.0% (77/77)	100.0%
Heteroplasmic (10-15%)	100.0% (74/74)	100.0%
Heteroplasmic (5-10%)	100.0% (3/3)	100.0%
Heteroplasmic (<5%)	50.0% (2/4)	100.0%
CLINICAL VALIDATION (n=76 samples)		
All types		
Single nucleotide variants n=2084 SNVs		
Heteroplasmic (45-100%)	100.0% (1940/1940)	100.0%
Heteroplasmic (35-45%)	100.0% (4/4)	100.0%
Heteroplasmic (25-35%)	100.0% (3/3)	100.0%
Heteroplasmic (15-25%)	100.0% (3/3)	100.0%
Heteroplasmic (10-15%)	100.0% (9/9)	100.0%
Heteroplasmic (5-10%)	92.3%(12/13)	99.98%
Heteroplasmic (<5%)	88.7% (47/53)	99.93%
Insertions and deletions by sequence analysis n=42 indels		
Heteroplasmic (45-100%) 1-10bp	100.0% (32/32)	100.0%
Heteroplasmic (5-45%) 1-10bp	100.0% (3/3)	100.0%
Heteroplasmic (<5%) 1-10bp	100.0% (5/5)	>0.9999
SIMULATION DATA /(mitomap mutations)		
Insertions, and deletions 1-24 bps by sequence analysis; n=17		
Homoplasmic (100%) 1-24bp	100.0% (17/17)	99.98%
Heteroplasmic (50%)	100.0% (17/17)	99.99%

Heteroplasmic (25%)	100.0% (17/17)	100.0%
Heteroplasmic (20%)	100.0% (17/17)	100.0%
Heteroplasmic (15%)	100.0% (17/17)	100.0%
Heteroplasmic (10%)	94.1% (16/17)	100.0%
Heteroplasmic (5%)	94.1% (16/17)	100.0%
Copy number variants (separate artifical mutations; n=1500)		
Homoplasmic (100%) 500 bp, 1kb, 5 kb	100.0%	100.0%
Heteroplasmic (50%) 500 bp, 1kb, 5 kb	100.0%	100.0%
Heteroplasmic (30%) 500 bp, 1kb, 5 kb	100.0%	100.0%
Heteroplasmic (20%) 500 bp, 1kb, 5 kb	99.7%	100.0%
Heteroplasmic (10%) 500 bp, 1kb, 5 kb	99.0%	100.0%
The performance presented above reached by following coverage metrics at assay level (n=66)		
	Mean of medians	Median of medians
Mean sequencing depth MQ0 (clinical)	18224X	17366X
Nucleotides with >1000x MQ0 sequencing coverage (%) (clinical)	100%	
rho zero cell line (=no mtDNA), mean sequencing depth	12X	

Bioinformatics

The target region for each gene includes coding exons and ±20 base pairs from the exon-intron boundary. In addition, the panel includes non-coding variants if listed above (Non-coding variants covered by the panel). Some regions of the gene(s) may be removed from the panel if specifically mentioned in the 'Test limitations" section above. The sequencing data generated in our laboratory is analyzed with our proprietary data analysis and annotation pipeline, integrating state-of-the art algorithms and industry-standard software solutions. Incorporation of rigorous quality control steps throughout the workflow of the pipeline ensures the consistency, validity and accuracy of results. Our pipeline is streamlined to maximize sensitivity without sacrificing specificity. We have incorporated a number of reference population databases and mutation databases such as, but not limited, to 1000 Genomes Project, gnomAD, ClinVar and HGMD into our clinical interpretation software to make the process effective and efficient. For missense variants, *in silico* variant prediction tools such as SIFT, PolyPhen, MutationTaster are used to assist with variant classification. Through our online ordering and statement reporting system, Nucleus, the customer has an access to details of the analysis, including patient specific sequencing metrics, a gene level coverage plot and a list of regions with inadequate coverage if present. This reflects our mission to build fully transparent diagnostics where customers have easy access to crucial details of the analysis process.

Clinical interpretation

We provide customers with the most comprehensive clinical report available on the market. Clinical interpretation requires a fundamental understanding of clinical genetics and genetic principles. At Blueprint Genetics, our PhD molecular geneticists, medical geneticists and clinical consultants prepare the clinical statement together by evaluating the identified variants in the context of the phenotypic information provided in the requisition form. Our goal is to provide clinically meaningful statements that are understandable for all medical professionals regardless of whether they have formal training in genetics.

Variant classification is the corner stone of clinical interpretation and resulting patient management decisions. Our classifications follow the Blueprint Genetics Variant Classification Schemes based on the ACMG guideline 2015. Minor modifications were made to increase reproducibility of the variant classification and improve the clinical validity of the report. Our experience with tens of thousands of clinical cases analyzed at our laboratory allowed us to further develop the industry standard.

The final step in the analysis of sequence variants is confirmation of variants classified as pathogenic or likely pathogenic using bi-directional Sanger sequencing. Variant(s) fulfilling the following criteria are not Sanger confirmed: the variant quality score is above the internal threshold for a true positive call, and visual check-up of the variant at IGV is in-line with the variant call. Reported variants of uncertain significance are confirmed with bi-directional Sanger sequencing only if the quality score is below our internally defined quality score for true positive call. Reported copy number variations with a size <10 exons are confirmed by orthogonal methods such as qPCR if the specific CNV has been seen less than three times at Blueprint Genetics.

Our clinical statement includes tables for sequencing and copy number variants that include basic variant information (genomic coordinates, HGVS nomenclature, zygosity, allele frequencies, in silico predictions, OMIM phenotypes and classification of the variant). In addition, the statement includes detailed descriptions of the variant, gene and phenotype(s) including the role of the specific gene in human disease, the mutation profile, information about the gene's variation in population cohorts and detailed information about related phenotypes. We also provide links to the references used, congress abstracts and mutation variant databases used to help our customers further evaluate the reported findings if desired. The conclusion summarizes all of the existing information and provides our rationale for the classification of the variant.

Identification of pathogenic or likely pathogenic variants in dominant disorders or their combinations in different alleles in recessive disorders are considered molecular confirmation of the clinical diagnosis. In these cases, family member testing can be used for risk stratification within the family. In the case of variants of uncertain significance (VUS), we do not recommend family member risk stratification based on the VUS result. Furthermore, in the case of VUS, we do not recommend the use of genetic information in patient management or genetic counseling.

Our interpretation team analyzes millions of variants from thousands of individuals with rare diseases. Thus, our database, and our understanding of variants and related phenotypes, is growing by leaps and bounds. Our laboratory is therefore well positioned to re-classify previously reported variants as new information becomes available. If a variant previously reported by Blueprint Genetics is re-classified, our laboratory will issue a follow-up statement to the original ordering health care provider at no additional cost.

#}

ICD codes

Commonly used ICD-10 codes when ordering the Purine and Pyrimidine Metabolism Disorders Panel

ICD-10	Disease
E79.1	Lesch-Nyhan syndrome
D81.5	Purine nucleoside phosphorylase deficiency
D81.3	Severe combined immunodeficiency due to adenosine deaminase deficiency

Accepted sample types

- EDTA blood, min. 1 ml
- Purified DNA, min. 3µg*
- Saliva (Oragene DNA OG-500 kit)

Label the sample tube with your patient's name, date of birth and the date of sample collection.

Note that we do not accept DNA samples isolated from formalin-fixed paraffin-embedded (FFPE) tissue.

Resources

- GeneReviews- Adenine Phosphoribosyltransferase Deficiency
- GeneReviews- Adenosine Deaminase Deficiency
- GeneReviews- Autosomal Dominant Tubulointerstitial Kidney Disease, UMOD-Related
- GeneReviews- Lesch-Nyhan Syndrome
- GeneReviews- Phosphoribosylpyrophosphate Synthetase Superactivity