ARUP LABORATORIES | NATIONAL REFERENCE LABORATORY

Exome Sequencing

Mendelian diseases are inherited conditions linked to individual genes. This test entails whole exome sequencing of a patient (and both parents, if available) with a suspected Mendelian genetic disorder to identify potentially pathogenic variants in ~19,000 genes of known and unknown function.

TEST OVERVIEW

Parental specimens are required to identify de novo variants, identify chromosomal phase of variants and interpret patient results.

- The exome accounts for 1-2% of the human genome but harbors ~85% of pathogenic variants.
- The function of only ~4,500 genes is currently known.
- Exome sequencing may or may not
  - Determine etiology of patient's medical condition
  - Predict prognosis or severity
  - Guide medical management

Information Required for Testing

- Completed Informed Consent for Exome Sequencing form for patient and parents
- Completed Patient History for Exome Sequencing form for proband and parents
- Three-generation medical pedigree
- Results of genomic microarray, magnetic resonance imaging (MRI) and other relevant tests
- Summary notes from genetic and specialist consultations

TEST DESCRIPTION

Clinical Sensitivity

Unpublished internal data

- ~20% when only the proband is sequenced and one or both parental samples are unavailable
- ~35% when only performing targeted sequencing of parental samples based on variants identified by sequencing the proband's exome
- ~45% when the proband and both parents undergo exome sequencing

Reporting

- Tens of thousands of genetic variants are identified.
  - Variants may be pathogenic, benign, or of unknown clinical significance.
  - American College of Medical Genetics (ACMG) recommends reporting secondary findings in genes not associated with the patient's clinical phenotype. (See table below.)
- Only the following variants are reported:
  - Those predicted to be related to the patient's phenotype
  - De novo and rare compound heterozygous variants in genes of unknown function, if a causative variant is not identified

TESTS TO CONSIDER

Exome Sequencing, Trio 2006332
Method: Massively Parallel Sequencing

Exome Sequencing, Familial Control 2006340
Method: Massively Parallel Sequencing
Exome Sequencing,Trio is the preferred test to diagnose an individual with an unknown Mendelian condition.
Order control test on patient's parents and up to two additional relatives who are affected with the same condition as the patient. It is performed at no additional cost and increases the detection rate.

Exome Sequencing, Proband 2006336
Method: Massively Parallel Sequencing

Exome Control, Targeted Sequencing 3001114
Method: Targeted Sanger Sequencing
The Exome Sequencing Proband test should only be ordered when it is not possible to obtain control samples from both biological parents.
Order Exome Control, Targeted Sequencing on patient's parents and additional relatives who are affected with the same condition as the patient. It is performed at no additional cost and increases the detection rate.

Additional Methodology

- Short tandem repeats (STRs) used to confirm familial relationships
- Liquid RNA- or DNA-based probes capture exons and intron/exon junctions of known protein-coding RefSeq genes
- Human reference sequence (Hg19) used for variant identification
- Variants confirmed by Sanger sequencing as needed

- Pathogenic variants in genes recommended by ACMG, or other medically actionable incidental variants in non-ACMG genes if elected on the consent form

- Single disease-causing variants in autosomal recessive ACMG genes are not reported.
- Family members undergoing exome sequencing who complete a consent form requesting ACMG variants will receive a separate report describing pathogenic secondary findings.
- Results are reported in 4-8 weeks.

- Additional testing, such as X-chromosome inactivation, may be performed to aid variant interpretation

Interpretation/Storage/Reanalysis/Data Sharing

- Accurate representation of biological relationships between family members is imperative for correct test interpretation.
- Test interpretation is based on information available at the time of testing and may change in the future.
- Exome sequencing data will be stored for a minimum of 5 years in compliance with ARUP's data retention policy, but many samples are discarded after testing is complete.
- Samples may be stored indefinitely for test validation or education purposes after personal identifiers are removed. All New York samples are discarded 60 days following test completion. Patients may request disposal of their sample by calling ARUP Laboratories at (800) 242-2787 x3301.
- The first request for reanalysis of data is available at no cost; subsequent reanalysis requests will be associated with charges.
- De-identified information about genetic variants and clinical findings may be published in international databases unless declined on consent form.
- Patients may also contact ARUP Laboratories at (800) 242-2787, ext. 3301, and request that their test result not be shared with public databases.
- Patients have the opportunity to participate in patient registries and research. For more information, see www.aruplab.com/genetics/resources.
- Raw exome sequencing data may be requested by the ordering healthcare provider and hospital that submitted the test to ARUP.

Limitations

- A negative result does not exclude a genetic cause for the patient's disorder.
- Diagnostic errors can occur due to rare sequence variations.
- Result interpretation may be impacted if this individual has had an allogeneic stem cell transplantation.
- The human exome is not able to be completely analyzed as some genes have not been identified while others, due to technical limitations, cannot either be sequenced or interpreted.
- The following will not be evaluated:
  - Variants outside the coding regions and intron-exon boundaries of the targeted genes
  - Regulatory region variants and deep intronic variants
  - Large deletions/duplications
  - Mitochondrial DNA (mtDNA)
- The following may not be detected:
  - Deletions/duplications/insertions of any size by massively parallel sequencing
  - Some variants due to the presence of pseudogenes, repetitive, or homologous regions
  - Low-level somatic variants
  - Chromosomal phase of identified variants
  - Some of the pathogenic ACMG variants that cannot be detected by routine exome analysis

Analytical Sensitivity

The analytical sensitivity of this test is approximately 98% for single nucleotide variants (SNVs) and greater than 93% for insertions/duplications/deletions from 1-10 base pairs in size. Deletions/duplication greater than 10 base pairs may be detected, but the analytical sensitivity may be reduced.

ACMG (Kalia, 2016) Recommends Reporting Secondary Findings for These Genes

Conditions		Associated Genes
Tumors/cancer syndromes	Familial adenomatous polyposis	*APC*
	Familial medullary thyroid cancer Multiple endocrine neoplasia type 2	*RET*
	Hereditary breast and ovarian cancer	*BRCA1, BRCA2*

Conditions		Associated Genes
	Hereditary paraganglioma/pheochromocytoma	*SDHD, SDHAF2, SDHC, SDHB*
	Juvenile polyposis	*BMPR1A*, *SMAD4*
	Li-Fraumeni syndrome	*TP53*
	Lynch syndrome	*MLH1, MSH2, MSH6, PMS2*
	Multiple endocrine neoplasia type 1	*MEN1*
	MUTYH-associated polyposis	*MUTYH*
	Neurofibromatosis type 2	*NF2*
	Peutz-Jeghers syndrome	*STK11*
	PTEN hamartoma tumor syndrome	*PTEN*
	Retinoblastoma	*RB1*
	Tuberous sclerosis complex	*TSC1, TSC2*
	Von Hippel-Lindau syndrome	*VHL*
	WT1-related Wilms tumor	*WT1*
Cardiovascular conditions/syndromes	Arrhythmogenic right-ventricular cardiomyopathy	*PKP2, DSP, DSC2, TMEM43, DSG2*
	Brugada syndrome Romano-Ward long QT syndrome types 1, 2, and 3	*KCNQ1, KCNH2, SCN5A*
	Catecholaminergic polymorphic ventricular tachycardia	*RYR2*
	Ehlers-Danlos syndrome, vascular type	*COL3A1*
	Familial hypercholesterolemia	*LDLR, APOB, PCSK9*
	Familial thoracic aortic aneurysms and dissections	*SMAD3, ACTA2, MYLK, MYH11*
	Hypertrophic cardiomyopathy, dilated cardiomyopathy	*MYBPC3, MYH7, TNNT2, TNNI3, TPM1, MYL3, ACTC1, PRKAG2, GLA, MYL2, LMNA*
	Loeys-Dietz	*TGFBR1*, *TGFBR2*
	Marfan syndrome	*FBN1*
Other conditions	Malignant hyperthermia susceptibility	*RYR1, CACNA1S*
	Ornithine transcarbamylase deficiency	*OTC*
	Wilson disease	*ATP7B*

REFERENCES

Farwell KD, Shahmirzadi L, El-Khechen D, Powis Z, Chao EC, Davis BT, Baxter RM, Zeng W, Mroske C, Parra MC, Gandomi SK, Lu I, Li X, Lu H, Lu H, Salvador D, Ruble D, Lao M, Fischbach S, Wen J, Lee S, Elliott A, Dunlop CL, Tang S. Enhanced utility of family-centered diagnostic exome sequencing with inheritance model-based analysis: results from 500 unselected families with undiagnosed genetic conditions Genet Med. 2015; 17(7): 578-86. PubMed

Kalia SS, Adelman K, Bale SJ, Chung WK, Eng C, Evans JP, Herman GE, Hufnagel SB, Klein TE, Korf BR, McKelvey KD, Ormond KE, Richards S, Vlangos CN, Watson M, Martin CL, Miller DT. Recommendations for reporting of secondary findings in clinical exome and genome sequencing, 2016 update (ACMG SF v2.0): a policy statement of the American College of Medical Genetics and Genomics Genet Med. 2016; PubMed

ARUP Laboratories is a nonprofit enterprise of the University of Utah and its Department of Pathology.
500 Chipeta Way, Salt Lake City, UT 84108 | (800) 522-2787 | (801) 583-2787 | aruplab.com | arupconsult.com
Content Review November 2018 | Last Update February 2019