

ATP7B Gene Analysis in Wilson Disease

Clinical Features:

Wilson disease is an autosomal recessive disorder characterized by excessive copper accumulation in various organs, primarily the liver, brain, kidney and cornea and can present with hepatic, neurologic or psychiatric abnormalities or a combination of all these. The clinical course is highly heterogeneous and age of onset ranges from 2 years to over 70 years.¹ Liver disease is commonly the first symptom in children and younger adults and is very rarely accompanied by neurologic or psychiatric symptoms, while neurologic/psychiatric symptoms are usually presenting symptoms in adults and can be present with or without liver findings.⁴ Kayser-Fleisher rings, resulting from copper deposition in the cornea, are considered a hallmark finding in patients with Wilson disease and are present in approximately 90%-95% of patients presenting with neurologic or psychiatric symptoms and in over half of those without these symptoms. However, in children presenting with liver disease Kayser-Fleischer rings are usually absent.^{2,3} Liver findings are highly variable ranging from abnormalities on liver function tests to acute liver failure, acute hepatitis or liver cirrhosis.⁴ Fatty liver, chronic liver disease and hemolytic anemia may also be present.³ Neurologic findings include tremors, poor coordination, loss of fine-motor control, chorea, choreoathetosis, and spastic dystonia, while psychiatric findings include depression, neurotic behavior, mood disturbance and intellectual deterioration.³ Other symptoms include renal involvement, arthritis, reduced bone mineral density, pancreatitis, cardiomyopathy, cardiac arrhythmias, rhabdomyolysis, endocrine disorders and sunflower cataracts.³ If untreated, Wilson disease can be lethal; however, symptoms can be prevented if affected patients are diagnosed and treated early.⁵

Genetics and Biochemical Features:

Wilson disease is caused by pathogenic variants in the *ATP7B* gene that is located on chromosome 13q14 and encodes a copper-transporting P-type ATPase, which delivers copper for incorporation into apoceruloplasmin and excretion into the bile. Impaired *ATP7B* function results in excessive cellular copper accumulation and clinical symptoms. A diagnosis of Wilson disease is based on a combination of clinical and laboratory findings. Biochemical tests include liver function tests, liver biopsy findings, liver copper concentration, serum ceruloplasmin concentration and 24-hour urinary copper concentration. Ultimately, confirmation of a diagnosis depends on the identification of disease-causing variants.⁶ The incidence of Wilson disease is estimated to be 1 in 30,000 individuals although the prevalence has been reported to be higher in certain regions and ethnic groups.⁵

Test Methods:

Using genomic DNA extracted from the submitted specimen, the complete coding regions and splice site junctions, as well as a promoter region encompassing the c.-436_-422del15 variant, of the *ATP7B* gene 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 the reference sequence 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; however, technical limitations and inherent sequence properties effectively reduce this resolution for some genes. Alternative sequencing or copy number detection methods are used to analyze or confirm regions with inadequate sequence or copy number data by NGS. 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. The copy number assessment methods used with this test cannot reliably detect copy number variants of less than 500 base pairs or 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 Sensitivity:

In a number of studies including large numbers of patients with Wilson disease from different ethnic backgrounds, who met clinical diagnostic standards, sequencing of the *ATP7B* gene identified reportedly pathogenic variants on both alleles in 88% to 90% of patients.^{4,5} When multiplex ligation-dependent probe amplification (MLPA) testing was also performed, the molecular diagnostic rate increased by 3%.⁸

Variant Spectrum:

At this time there are over 900 variants in the *ATP7B* gene associated with Wilson disease in the Human Gene Mutation Database. The vast majority are missense, with nonsense, splicing, regulatory, small deletions/insertions and large deletions also reported. In 168 Chinese patients, exon-level large deletions of *ATP7B* were identified on 6 alleles.⁸ At this time, there is poor genotype-phenotype correlation for Wilson disease.⁹ However, individuals homozygous for the p.H1069Q variant, which accounts for 35%-45% of Wilson disease alleles in a mixed European population, were reported to have a mean age of onset of 20 to 22 years.³ Other variants common to specific ethnic groups include p.R778L, accounting for approximately 57% of disease alleles in the Asian population younger than 18 years and a 15 bp deletion in the promotor region which is common in Sardinia.³

References:

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