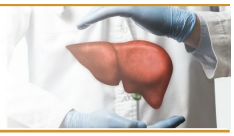
THE PATHCARE NEWS

GENETIC TESTING FOR CHILDHOOD LIVER DISORDERS



When evaluating a patient with abnormal liver functions, it is important to investigate the potential genetic causes.

1. Hyperbilirubinemia

Neonatal hyperbilirubinemia results from elevated total serum bilirubin (TSB). In most cases, it is a mild, transient condition; however, it is imperative to distinguish this from a more severe form called "pathological jaundice". Conditions causing hyperbilirubinemia can be classified into those that result in a predominantly unconjugated hyperbilirubinemia and those that are associated with the conjugated form of bilirubin.

• Unconjugated hyperbilirubinemia

Unconjugated hyperbilirubinemia arises in one of the three major pathophysiologic conditions or a combination of them:

- Increased bilirubin production
- Impaired bilirubin uptake
- Impaired bilirubin conjugation

Hereditary defects causing impaired bilirubin conjugation:

- **Gilbert syndrome**
- **Crigler-Najjar syndrome type I and II**

	Gilbert syndrome	Crigler- Najjar syndrome 1	Crigler-Najjar syndrome 2
Gene responsible	*UGT1A1 Promotor variant*	*UGT1A1*	*UGT1A1*
Inheritance pattern	AR* (mostly)	AR	AR
% residual enzyme	25%	undetectable	partial deficiency
Total serum bilirubin levels	1-6 mg/dL	20 to 45 mg/dL	6 to 20 mg/dL
Clinically	Mild self-limiting	Severe; Kernicterus	Less severely jaundiced

** Autosomal recessive*

• Conjugated hyperbilirubinemia or cholestasis

The causes of neonatal cholestasis are extensive and can be classified into the following categories:

- Obstruction of biliary flow
- Infections
- Genetic causes
  - **Cystic fibrosis**
  - **Alpha-1 anti-trypsin deficiency**
  - Other - Alagille syndrome, galactosemia, fructosemia, tyrosinemia type 1, progressive familial intrahepatic cholestasis (PFIC), Aagenaes syndrome, Dubin-Johnson syndrome, Bile acid synthesis disorders, etc.

Cystic fibrosis

Cystic fibrosis (CF) is an inherited genetic condition that primarily affects the lungs and pancreas by clogging them with thick and sticky mucus. It is caused by genetic variants in the cystic fibrosis transmembrane conductance regulator (*CFTR*) gene. CF follows an autosomal recessive inheritance pattern.

The carrier frequency of CF in Caucasian population in South Africa is 1 in 23 with the delta F508 variant being the most common variant amongst Caucasians (prevalence 76%), whereas 3120+1G>A is the most common mutation (prevalence 46%) in black Africans, with an estimated carrier frequency rate of 1 in 90 healthy individuals.[1]

Alpha-1-antitrypsin deficiency

Alpha-1 antitrypsin deficiency (AATD) is a genetic condition that increases the risk of lung and liver disease. It is caused by genetic variants in the *SERPINA1* gene that results in the production of an abnormal type of the Alpha-1 protein. Individuals with AATD have both copies of their *SERPINA1* gene mutated. There are multiple variants of the alpha-1 antitrypsin genotype, with some of these variants producing normal levels of alpha-1 antitrypsin and some with reduce levels (Table below). The reduced levels are associated with disease.

SERPINA1 allele	Description
PI*M	Most common allele in all populations
PI*Z	Most common pathogenic allele. Results in a quantitatively and functionally deficient AAT protein. Individuals homozygous for PI*Z (i.e., PI*ZZ) have severe AATD
PI*S	Pathogenic allele resulting in a quantitatively and functionally deficient AAT. Only clinically significant in the compound heterozygous state with another pathogenic allele (e.g., PI*SZ) and when the serum AAT level is <57 mg/dL.
PI*F	Pathogenic allele. Resulting protein is functionally impaired in binding neutrophil elastase but quantitatively normal.
PI*I	Allele associated with mild quantitative deficiency
PI*QO (null allele)	Pathogenic alleles that result in either no mRNA product or no protein production

2. Acute liver failure in children

Acute liver failure in infants and children can be caused by:

- Infections
- Drugs or toxins
- Cardiovascular conditions
- Immune related conditions
- Metabolic disorders
    - **Galactosemia, tyrosinemia, mitochondrial condition**, hereditary fructose intolerance, fatty acid oxidation, Reye's syndrome, leukemia, iron storage

Neonatal acute liver failure requires broad testing with a fast turnaround time. International testing options are available via Invitae and Prevention Genetics in the US, and Centogene in Germany (see Quick Guide below)

• Mitochondrial conditions

Mitochondrial disorders can be caused by:

1) Mitochondrial genome variants (maternally inherited)
2) Nuclear mitochondrial genes (mostly AR)

Local genetic testing includes:

- Mitochondrial point genetic variants
- Mitochondrial large deletion screen
- Full mitochondrial genome sequencing

• Newborn screening

Newborn screening test screens for a group of disorders called Inborn Errors of Metabolism (IEM), including **Tyrosinemia type 1** and **Galactosemia**, some endocrine disorders and Cystic Fibrosis.

PathCare has partnered with the Newborn Screening Laboratory of North-West University (NWU) and Next Biosciences to facilitate newborn screening for patients.

Invitae – International Send-away Service

PathCare offers a referral service to Invitae Laboratory in the USA. Invitae offers an extensive genetic test menu over a broad range of clinical areas.

Invitae offers testing via single-gene or multi-gene panels at a fixed patient-pay price, with the additional benefits of re-requisitioning a sample for additional genes in the same clinical area at no extra cost, and free-of-charge family variant testing to first-degree relatives, if requested within 150 days of the report. (A PathCare handling fee might be applicable for family members.)

The cost of any non-cancer Invitae diagnostic panel (regardless of the number of genes tested) is 349 US Dollars (cost in Rands is dependent on the exchange rate). As this is a patient-pay option, Invitae bills the patient directly for these tests. PathCare charges an international handling and courier fee (R950) which is paid upfront when providing the sample.

Blood samples are sent to Invitae in EDTA tubes. The turnaround time for Invitae tests is 3 – 4 weeks.

Centogene – International Send-away Service

PathCare also offers a referral service to Centogene Laboratory in Germany. Centogene offers NGS Hereditary Diagnostic Panels, as well as Whole exome sequencing (WES) and other test options. The cost of Centogene panels vary, however general costs are indicated below:

Solo Exome	630 USD
NGS Hereditary Diagnostic Panels	400 USD

PathCare charges a R750 international handling fee for this service. The turnaround time for Centogene tests is approximately 6 weeks for panels, and 7 - 8 weeks for exomes. A STAT option is available for WES at an additional fee with a turnaround time of 3 – 4 weeks.

Please feel free to contact our PathCare Genetics Team if you have any questions on 021 596 3655 or geneticconsult@pathcare.org

References

1. Zampoli, M., Verstraete, J., Frauendorf, M., Kassanjee, R., Workman, L., Morrow, B. M., & Zar, H. J. (2021). Cystic fibrosis in South Africa: spectrum of disease and determinants of outcome. ERJ open research, 7(3), 00856-2020. https://doi.org/10.1183/23120541.00856-2020

Quick Guide to Testing

Local testing offered through PathCare					
Condition/ Syndrome	**Gilbert syndrome**	**Alpha-1 antitrypsin deficiency (AATD)**	**Galactosemia**	**Mitochondrial testing**	**Cystic fibrosis**
Description	a. *UGT1A1* promotor variant *(Gilbert syndrome)* b. *UGT1A1* full sequencing *(Crigler-Najjar syndrome)*	a. Antitrypsin enzyme analysis b. AATD (*SERPINA1*: S and Z alleles only)	*GALT*: common SA variant c.404C>T	a. Full MtDNA genome sequencing b. Large deletion screen c. Mitochondrial point genetic variants	*CFTR* gene a. Delta F508 variant analysis b. *CFTR* full sequencing

International testing offered through PathCare							
Laboratory	**Centogene**		**Invitae**				**Prevention Genetics**
Location	**Germany**		**United States of America**				**United States of America**
Condition/ Syndrome	Mitochondrial testing	Neonatal acute liver failure	Crigler Najjar & Gilbert syndrome	Neonatal acute liver failure	Conjugated hyperbiliru-binemia	Cystic Fibrosis	Neonatal acute liver failure
Description	Full mitochondrial genome sequencing and nuclear mitochondrial gene sequencing	Whole exome sequencing (incl. mitochondrial genome analysis) (Solo, Trio & STAT)	*UGT1A1* gene	Panels: • Cholestasis • Newborn Screening confirmation • Supplemental Newborn Screening • Nuclear mitochondrial disorders • Hereditary Hemophagocytic Lymphohistiocytosis (HLH) Disorders • Fatty acid oxidation defects	Cholestasis panel	*CFTR* full gene sequencing	Neonatal Crisis panel (Family Trio vs Patient only options)
Turnaround time	~ 6 weeks	Solo/Trio: ~7-8 weeks STAT: ~3 - 4 weeks	~3 – 4 weeks				~ 3 weeks
Add on extras			Free family testing (within 150 days) – PathCare handling fee applicable for family members outside of household Free of charge re-requisition (within 150 days), if the initial test is negative – within same clinical area				

** Pricing is correct and valid for 2023. Costs are subject to change. Please contact the PathCare genetics team for confirmation of current prices.*