# Whole Genome Sequencing
## *Couple Carrier Sequencing*

### Test Description
Whole exome sequencing covers 20321 genes including 37 mitochondria genes. Couple carrier screening is useful in recessive disorders to find whether you or/ and your partner carry the genetic defect that could cause an inherited genetic disorder in your baby. If both the partners are carriers for a specific inherited recessive disorder, they would have a 1 in 4 (25%) chance in each pregnancy, of having a baby with that recessive disease.

### Patient Demographic
**Name**: Mr. Hardeep Banga
**Sex**: Male
**Date of Birth/Age**: 59 years
**Disease**: Heart problem, neurological problem

### Clinician
Clinician Name: Dr Amit Verma
Medical Facility: Dr AV Institute of Personalized Therapy and Cancer Research (IPTCR)
Pathologist: Not Provided

### Specimen
**Booking ID**: 012311090055
**Site**: NA
**Sample Type**: Blood
**Date of Collection**: 09-11-2023
**Date of Booking**: 09-11-2023

## CLINICAL SYNOPSIS
Hardeep Banga has clinical indications of heart problem, neurological problem and h/o abortion. Couple carrier sequencing was performed.

## RESULTS

**Pathogenic (carrier) variant detected.**

| Gene (Transcript) # | Location | Variant & Depth/VAF | Zygosity | Disease (OMIM/Ref) | Inheritance | Classification |
| --- | --- | --- | --- | --- | --- | --- |
| *TIMM22 (+)* NM_013337.4 (chr17:g.997217C>A) | Exon 1 | c.75C>A (p.Tyr25Ter) 64x/43% | Heterozygous | ? Combined oxidative phosphorylation deficiency 43 | Autosomal recessive | Pathogenic (PVS1, PP5, PM2) |
| *MYLK (-)* NM_053025.4 (chr3:g.123707753C>T) | Intron 16 | c.2390+1G>A (Splice donor variant) 42x/52% | Heterozygous | Megacystis-microcolon-intestinal hypoperistalsis syndrome 1 | Autosomal recessive | Likely pathogenic (PVS1, PM2) |
| *CFTR (+)* NM_000492.4 (chr7:g.117548798T>C) | Exon 10 | c.1367T>C (p.Val456Ala) 92x/48% | Heterozygous | Cystic fibrosis | Autosomal recessive | Pathogenic (PM1, PP3, PS1, PM5, PM2) |
| *IFT140 (-)* NM_014714.4 (chr16:g.1586129C>T) | Intron 10 | c.1155+1G>A (Splice donor variant) 31x/38% | Heterozygous | Short-rib thoracic dysplasia 9 with or without polydactyly | Autosomal recessive | Pathogenic (PM2, PVS1, PP5) |

**Genetic test results are reported based on the recommendations of American College of Medical Genetics*

## ? COMBINED OXIDATIVE PHOSPHORYLATION DEFICIENCY 43

***OMIM phenotype***: Pacheu-Grau *et al*. (2018) reported a 10-year-old British girl who presented at birth with intrauterine growth retardation, hypotonia with decreased spontaneous movements, and feeding difficulties associated with gastroesophageal reflux. She continued to have poor overall growth and muscle weakness during childhood. Brain imaging showed delayed myelination, and laboratory studies showed increased serum lactate and creatine kinase. Additional clinical details were not provided. Muscle biopsy showed decreased activities of mitochondrial respiratory chain complexes I, III, and IV. Patient fibroblasts showed a partially fragmented mitochondrial network and a mild defect in oxygen consumption compared to controls. She had previously been reported by del Mar O'Callaghan *et al*. (2012).

## MEGACYSTIS-MICROCOLON-INTESTINAL HYPOPERISTALSIS SYNDROME 1

***OMIM phenotype***: Megacystis-microcolon-intestinal hypoperistalsis syndrome-1 (MMIHS1) is a congenital disorder characterized by loss of smooth muscle contraction in the bladder and intestine. A distended bladder (megacystis) may be detected on prenatal ultrasound. Intestinal malrotation has also been observed (summary by Halim *et al*., 2017).

## CYSTIC FIBROSIS

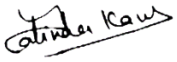
***OMIM phenotype***: Cystic fibrosis (CF) is classically described as a triad of chronic obstructive pulmonary disease, exocrine pancreatic insufficiency, and elevation of sodium and chloride concentration in sweat. Almost all males with CF are infertile due to congenital bilateral absence of the vas deferens. The disorder is associated with decreased longevity (summary by Cutting, 2002).

## SHORT-RIB THORACIC DYSPLASIA 9 WITH OR WITHOUT POLYDACTYLY

***OMIM phenotype***: Short-rib thoracic dysplasia (SRTD) with or without polydactyly refers to a group of autosomal recessive skeletal ciliopathies that are characterized by a constricted thoracic cage, short ribs, shortened tubular bones, and a 'trident' appearance of the acetabular roof. SRTD encompasses Ellis-van Creveld syndrome (EVC) and the disorders previously designated as Jeune syndrome or asphyxiating thoracic dystrophy (ATD), short rib-polydactyly syndrome (SRPS), and Mainzer-Saldino Syndrome (MZSDS). Polydactyly is variably present, and there is phenotypic overlap in the various forms of SRTDs, which differ by visceral malformation and metaphyseal appearance. Nonskeletal involvement can include cleft lip/palate as well as anomalies of major organs such as the brain, eye, heart, kidneys, liver, pancreas, intestines, and genitalia. Some forms of SRTD are lethal in the neonatal period due to respiratory insufficiency secondary to a severely restricted thoracic cage, whereas others are compatible with life (summary by Huber and Cormier-Daire, 2012 and Schmidts *et al*., 2013).

## RECOMMENDATIONS

- **Segregation analysis of the variant by Sanger sequencing is recommended in affected and unaffected members of the family.**
- Genetic counselling is advised for interpretation on the consequences of the variant(s).
- We recommend confirming the presence of these variants by Sanger Sequencing.
- The significance/classification of the variants might change based on parental and family members genetic testing.
- For questions about this report, or for assistance in locating nearby genetic counseling services, please contact the laboratory at contact@molq.in.
- If results obtained do not match the clinical findings, additional testing should be considered as per referring clinician's recommendations.



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## APPENDIX 1: TEST METHODOLOGY

### Method

**Next Generation Sequencing:**[1-5] DNA extracted from blood, saliva, amniotic fluid, CVS or any other standard source is used for targeted capture-based Library preparation. Targeted capture provides an efficient and sensitive means for sequencing specific genomic regions in a high-throughput manner. The libraries were sequenced to mean >85-100x coverage on Illumina Novaseq 6000 sequencing platform with Paired End 2x150 chemistry.

GATK best practices framework were followed for identification of variants in the sample. The sequences obtained are assembled and aligned to reference sequences based on NCBI RefSeq transcripts and human genome build (GRCh38). Haplotype caller has been used to identify variants which are relevant to the clinical indication.

In addition to SNVs and small Indels, copy number variants (CNVs) are detected from targeted sequence data using the commercially available algorithm. This algorithm detects rare CNVs based on comparison of the read-depths of the test data with the matched aggregate reference dataset. Clinically relevant mutations were annotated using published variants in literature, Commercial datasets and a set of diseases databases.

Common variants are filtered based on allele frequency in 1000Genome, ExAC, gnomAD, dbSNP and reference laboratory's internal database.

Non-synonymous variants effect is calculated using multiple algorithms such as PolyPhen-2, SIFT, MutationTaster2. Based on annotations data, ACMG rules-based classification performed for classification of variants identified through next generation sequencing study.

| Average Sequencing Depth (x) | Percentage Target Base Pairs Covered | | |
| --- | --- | --- | --- |
| | 1x | ≥5x | ≥20x |
| 79.58 | 99.09 | 98.92 | 98.52 |

| Title | Data |
| --- | --- |
| **Total data generated (Gb)** | 9.09 |
| **Total reads aligned (%)** | 100.00 |
| **Reads that passed alignment (%)** | 99.96 |
| **Data ≥ Q30 (%)** | 93.62 |

**$The classification of the variations is done based on American College of Medical Genetics as described below[6]**

| | |
| --- | --- |
| Variant | A change in a gene. This could be disease causing (pathogenic) or not disease causing (benign). |
| Pathogenic | A disease causing variation in a gene which can explain the patient's symptoms has been detected. This usually means that a suspected disorder for which testing had been requested has been confirmed. |
| Likely Pathogenic | A variant which is very likely to contribute to the development of disease however, the scientific evidence is currently insufficient to prove this conclusively. Additional evidence is expected to confirm this assertion of pathogenicity. |
| Variant of Uncertain Significance | A variant has been detected, but it is difficult to classify it as either pathogenic (disease causing) or benign (non-disease causing) based on current available scientific evidence. Further testing of the patient or family members as recommended by your clinician may be needed. It is probable that their significance can be assessed only with time, subject to availability of scientific evidence. |

### DISCLAIMER

- Interpretation of variants in this report is performed to the best knowledge of the laboratory based on the information available at the time of reporting. The classification of variants can change over time and MolQ cannot be held responsible for this. Please feel free to contact MolQ Laboratory (contact@molq.in) in the future to determine if there have been any changes in the classification of any variations. Re-analysis of variants in previously issued reports in light of new evidence is not routinely performed, but may be considered upon request.

- The sensitivity of this assay to detect large deletions/duplications of more than 10 bp or copy number variations (CNV) is 70-75%. The CNVs detected with this assay have to be confirmed by alternate method such as MLPA & Microarray.
- Due to inherent technology limitations, coverage is not uniform across all regions. Hence pathogenic variants present in areas of insufficient coverage as well as those variants which currently do not corelate with the provided phenotype may not be analyzed/ reported. Additionally, it may not be possible to fully resolve certain details about variants, such as mosaicism, phasing, or mapping ambiguity.
- Genes with pseudogenes, paralog genes and genes with low complexity may have decreased sensitivity and specificity of variant detection and interpretation due to inability of the data and analysis tools to unambiguously determine the origin of the sequence data in such regions.
- The mutations have not been validated/confirmed by Sanger sequencing.
- Incidental or secondary findings (if any) that meet the ACMG guidelines[2] can be given upon request.
- Sequence and copy number variants are reported according to the Human Genome Variation Society (HGVS).
- The transcript used for clinical reporting generally represents the canonical transcript, which is usually the longest coding transcript with strong/multiple supporting evidence. However, clinically relevant variants annotated in alternate complete coding transcripts could also be reported.
- The report shall be generated within turnaround time (TAT), however, such TAT may vary depending upon the complexity of test(s) requested. MolQ Laboratory under no circumstances will be liable for any delay beyond afore mentioned TAT.
- It is hereby clarified that the report(s) generated from the test(s) do not provide any diagnosis or opinion or recommends any cure in any manner. MolQ Laboratory hereby recommends the patient and/or the guardians of the patients, as the case may be, to take assistance of the clinician or a certified physician or doctor, to interpret the report(s) thus generated. MolQ Laboratory hereby disclaims all liability arising in connection with the report(s).
- In a very few cases genetic test may not show the correct results, e.g. because of the quality of the material provided to MolQ Laboratory. In case where any test provided by MolQ Laboratory fails for unforeseeable or unknown reasons that cannot be influenced by MolQ Laboratory in advance, MolQ Laboratory shall not be responsible for the incomplete, potentially misleading or even wrong result of any testing if such could not be recognized by MolQ Laboratory in advance.
- This is a laboratory developed test and the development and the performance characteristics of this test was determined by reference laboratory.

## LIMITATIONS

- Genetic testing is an important part of the diagnostic process. However, genetic tests may not always give a definitive answer. In some cases, testing may not identify a genetic variant even though one exists. This may be due to limitations in current medical knowledge or testing technology. Accurate interpretation of test results may require knowing the true biological relationships in a family. Failing to accurately state the biological relationships in {my/my child's} family may result in incorrect interpretation of results, incorrect diagnoses, and/or inconclusive test results.
- Test results are interpreted in the context of clinical findings, family history and other laboratory data. Only variations in genes potentially related to the proband's medical condition are reported. Rare polymorphisms may lead to false negative or positive results. Misinterpretation of results may occur if the information provided is inaccurate or incomplete.
- Triplet repeat expansions, translocations, large deletion or duplications and chromosomal rearrangements events are currently not reliably detected by next generation sequencing.
- This assay is not meant to interrogate most promoter regions, deep intronic regions, or other regulatory elements, other gene rearrangements like inversion or translocation and does not detect single or multiexon deletions or duplications.
- Genetic testing is highly accurate. Rarely, inaccurate results may occur for various reasons. These reasons include, but are not limited to: mislabelled samples, inaccurate reporting of clinical/medical information, rare technical errors or unusual circumstances such as bone marrow transplantation, blood transfusion; or the presence of change(s) in such a small percentage of cells that may not be detectable by the test (mosaicism).
- It should be noted that this test does not sequence all bases in a human genome, not all variants have been identified or interpreted, and this report is limited only to variants with evidence for causing or contributing to disease/clinical details provided to MolQ Laboratory.
- Testing has been performed assuming that the sample received belongs to the above named individual and any stated relationships between individuals are accepted as true.
- The results should be interpreted in the context of the patient's medical evaluation, family history and racial/ethnic background. Please note that variant classification and/or interpretation may change over time if more information available. Reinterpretation of multi gene next generation sequencing data is recommended on an annual basis and may be

requested by a medical provider.

## REFERENCES

1. Meyer L.R. et al., The UCSC Genome Browser database: extensions and updates 2013. Nucleic Acids Res., 41(D1):D64-9, 2013.
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4. Lek M. et al., Analysis of protein-coding genetic variation in 60,706 humans. Nature, 536(7616):285-91, 2016.
5. McLaren W. et al., Deriving the consequences of genomic variants with the Ensembl API and SNP Effect Predictor. Bioinformatics., 26(16):2069-70, 2010.
6. Richards S. *et al.*, Standards and guidelines for the interpretation of sequence variants: a joint consensus recommendation of the American College of Medical Genetics and Genomics and the Association for Molecular Pathology, Genet Med., 17(5):405-24, 2015.
7. Kalia S.S. et al., 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., 19(2):249-255, 2017.