Patient Name	Dummy	**Booking ID**	XXX
Age:	25 Years	**Sample Type**	Blood
Gender	Female	**Sample Collection Date**	22-10-2024
Reffering Doctor	Dr. XXX	**Sample Receiving Date**	23-10-2024
Test requested:	Beta thalassemia (HBB) deletion/ duplication analysis by MLPA	**Reporting Date**	08-11-2024

Beta thalassemia (*HBB*) deletion/ duplication analysis by MLPA

Clinical Indication/Symptoms
Referred for *HBB* gene deletion/duplication analysis via MLPA.

Test Result
NO PATHOGENIC/LIKELY PATHOGENIC DELETIONS/DUPLICATIONS CAUSATIVE OF THE SUSPECTED PHENOTYPE HAVE BEEN IDENTIFIED

Test result summary table

Deletion/Duplication	Gene (Exons) deleted/duplicated	MLPA Probe ratio (Final ratio)	Disease (omim)
NA	NA	NA	NA

Clinical Interpretation:
For this patient, No deletion/duplication was detected in abnormal range, within the detection limits of MLPA, in the *HBB* gene

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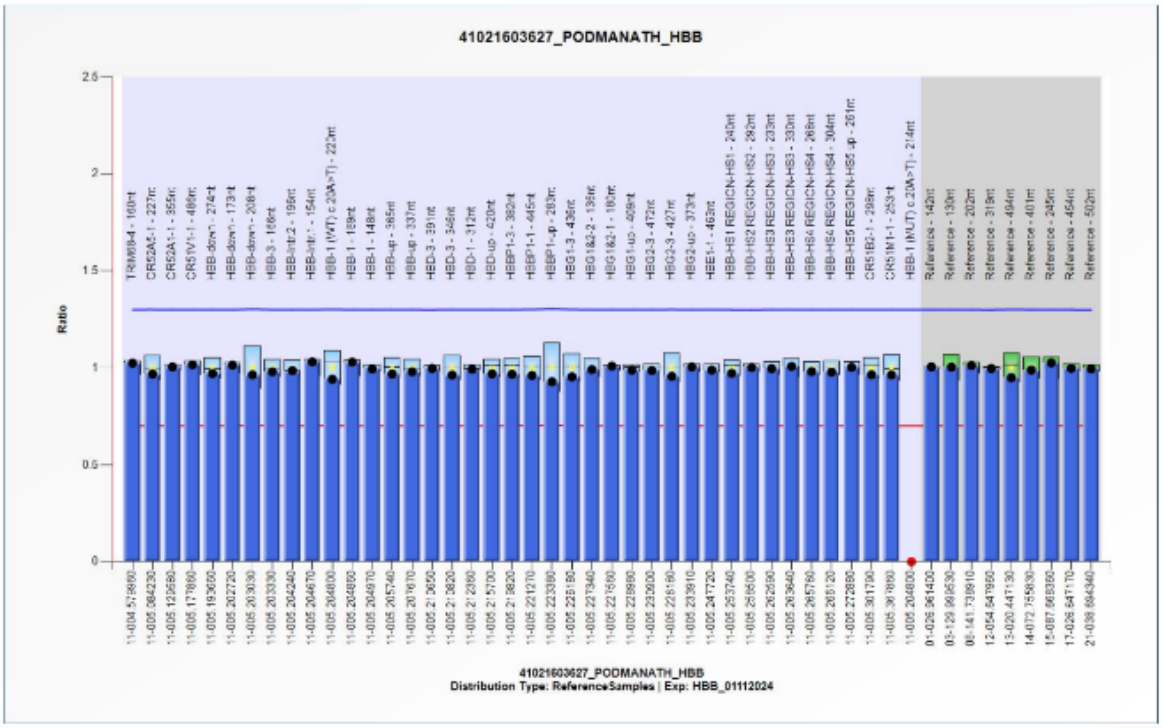

Comments:

- ✓ Please correlate clinically.
- ✓ Genetic counseling for accurate interpretation of test results is recommended.
- ✓ Gene sequencing or mutation analysis of the targeted region of the HBB gene is advised to rule out the presence of SNVs (single nucleotide variants) and INDELs (insertions and deletions).
- ✓ For further questions or queries regarding this report, please do not hesitate to contact Redcliffe Labs. Our team of experts is available to provide comprehensive explanations, discuss your results in detail, and offer guidance on the next steps.
- ✓ you can reach us through geneticcounselors@redcliffelabs.com.

Test Description

Beta thalassemia is an inherited blood disorder characterized by a reduced production of hemoglobin due to mutations in the HBB gene. This gene encodes beta-globin, a critical component of hemoglobin. Mutations in the HBB gene can lead to either a partial (β+) or complete (β0) absence of beta-globin, resulting in beta thalassemia and, in some cases, sickle cell anemia. In South-East Asia, around 20 different mutations—including deletions, insertions, base substitutions, and alternate splice variants—are linked to abnormal beta-globin production. In India, mutations such as del619bp, IVS1-5 G>C, IVS1-1 G>T, codon 8/9 (+G), and codon 41/42 (-TTCT) are notably prevalent. Genetic testing for the HBB gene can aid in diagnosing individuals with symptoms of beta thalassemia or related blood disorders. For parents with symptoms, a family history of the disorder, or known carrier status, prenatal testing can provide crucial information. Additionally, Multiplex Ligation-dependent Probe Amplification (MLPA) analysis is a valuable tool for detecting large deletions or duplications in the HBB gene, which can assist in confirming diagnoses and carrier status.

Methodology

Copy number changes in the beta-globin (HBB) gene cluster and its regulatory region on chromosome 11p15.4 can be identified using Multiplex Ligation-dependent Probe Amplification (MLPA) probes. MLPA probes consist of two hemi-probes that bind to adjacent sites on the target DNA sequence. When ligated and subsequently amplified by PCR, each unique MLPA probe generates an amplicon of distinct length, which is then separated and quantified using capillary electrophoresis.

Heterozygous deletions in the target sequences will reduce the efficiency of probe binding, resulting in a 35-50% decrease in the relative peak area of the amplification product for that specific probe set. By comparing MLPA peak patterns, copy number differences in various exons between test and control DNA samples can be detected.

Test Limitations

- ✓ The MLPA test will not detect the point mutations in HBB genes, which accounts for most of genetic defects. It is therefore recommended to use MLPA in combination with sequence analysis.
- ✓ A point mutation or polymorphism in the sequence detected by a probe, which results in reduced probe binding efficiency, can also cause a reduction in relative peak area. Therefore, single exon deletions detected by MLPA should always be confirmed by other methods like multiplex PCR or sequencing.
- ✓ MLPA cannot detect any changes that lie outside the target sequence of the probes and will not detect most inversions or translocations. Even when MLPA did not detect any aberrations, the possibility remains that biological changes in that gene or chromosomal region do exist but remain undetected.
- ✓ Although all precautions are taken during Molecular Genetic testing the currently available data indicate that the technical error rate for all types of Molecular DNA analysis is approximately 2%.

Disclaimer

- ✓ Interpretation of variants/CNV's 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 redcliffe labs cannot be held responsible for this. In the future to determine if there have been any changes in the classification of any variations. Please feel free to contact redcliffelabs.com
- ✓ The report shall be generated within turnaround time (TAT), however, such TAT may vary depending upon the complexity of test(s) requested 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. Redcliffe labs 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. Redcliffe labs 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 redcliffe labs . In case where any test provided by redcliffe labs fails for unforeseeable or unknown reasons that cannot be influenced by redcliffe labs in advance, redcliffe labs shall not be responsible for the incomplete, potentially misleading or even wrong result of any testing if such could not be recognized by redcliffe labs in advance.
- ✓ Redcliffe labs is not liable to provide diagnosis, opinion or recommendation related to disease, in any manner. Lab hereby recommends the Patient and/or the guardians of the Patient to contact clinician for further interpretation of the test results.
- ✓ This is a laboratory developed test and the development and the performance characteristics of this test was determined by redcliffe genomics labs

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- ✓ This test has not been validated by the FDA, NABL or CAP, and it has been determined by the accrediting bodies that such validation is not required at this time. **This report is for research purposes only, not for use in clinical diagnostic or therapeutic applications**.

Refrences:

- ✓ Galanello R1, Origa R. "Beta-thalassemia", Orphanet J Rare Dis. 2010 May 21;5:11
- ✓ Thein SL. "The molecular basis of β-thalassemia", Cold Spring Harb Perspect Med. 2013 May 1;3 (5):a011700.
- ✓ Dehury S, Purohit P, Meher S, Das K, Patel S. Compound heterozygous state of β-thalassemia with IVS1-5 (G→C) mutation and Indian deletion-inversion Gγ(Aγδβ)(0)-thalassemia in eastern India. Rev Bras Hematol Hemoter. 2015 May-Jun;37(3):202-6.

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