AWMGS
All Wales Medical Genomics Service

Test Information Sheet - Wales Infants' and ChildreN's Genome Service - WINGS

The Rapid Whole Genome (WGS) Sequencing Service for acutely unwell children with a likely monogenic disorder **(R14)** is available from April 2020.

Description

AWMGS Genetic Consultants, Bioinformaticians and Clinical Scientists have been working in partnership with specialist Paediatric Consultants and Cardiff University to develop the UK's first diagnostic rapid Whole Genome Sequencing (WGS) service for paediatric patients. This service will be available from April 2020 for patients being cared for in the neonatal or paediatric intensive care units who have a combination of serious health problems. The AWMGS WINGS test is designed to reduce the child's diagnostic journey by searching through their whole genome data to find a genetic cause of their clinical features.

Research studies to examine the application of WGS in a routine clinical setting have shown that it is highly effective in improving outcomes for this group of patients by reducing the need for multiple diagnostic tests, facilitating earlier treatment decisions and preventing repeat inpatient admissions (Kingsmore et al., 2019, PMID: 31564432; French et al. 2019, PMID: 30847515; Mestek-Boukhibar et al. 2018, PMID: 30049826).

Referral Criteria

Rapid whole genome sequencing (WGS) is available for acutely unwell children with a likely underlying genetic cause (R14). This test is only available on a trio basis and the child and BOTH parents must be available. Alternative rapid genetic testing is available if only one parent is present.

For full details, please see the referral criteria leaflet for this service (MI-GEN-RWGSRefCrit).

Result Reporting and Risk of Incidental findings

It is possible that as part of this test, incidental or secondary findings that are not of direct relevance to the clinical features in the patient may be identified. These **will not** be reported in the patient's initial result.

If the patient or family wish to receive information about additional, clinically actionable findings unrelated to the patient's clinical features, they can discuss this during a follow-up appointment with Clinical Genetics.

Trio whole genome sequencing will reveal possible non-paternity (or non-maternity) and this result would be discussed with the referring clinician.

If a diagnosis is not found, the variant data from genome sequencing tests will be stored long-term in order that further analysis can be undertaken in the future. The results of re-analyses may be issued as a supplementary report to the referring clinician.

Test Methods

Genomic DNA extracted from the child's and parent's blood samples is sequenced simultaneously using next generation sequencing (NGS) with the Illumina NovaSeq 6000 system. Sequences are aligned to human genome assembly GRCh37 (hg19), and variants identified with the Illumina DRAGEN (Dynamic Read Analysis for GENomics) Bio-IT Platform (v.3.7, Illumina).

Analysis includes evaluation of variants that are identified to be de novo, compound heterozygous, homozygous and X-linked using in-house bioinformatic pipelines. Variants are interpreted and reported following the latest ACGS/ACMG guidelines (Ellard et al., 2020; Richards et al., 2015).

Limitations

Currently this test is validated for the detection of small variants (SNV and Indels) from whole genome sequencing data only. This analysis does not include structural genetic variants such as inversions or translocations, copy number variants, mitochondrial variants, triplet repeats or UPD. This assay may not detect multi-nucleotide variants. Array CGH testing will be carried out concurrently for an interim period.

The rWGS analysis is primarily looking for de novo variation seen in offspring but not parents. Therefore, variants inherited from a parent with milder clinical features or disorders with a later age of onset may not be detected.

This test will not detect mosaicism for SNV or Indel variants.

Intronic variants: only conserved splice site variants (defined as variants located within 1–3 bases of an exon or 1–8 bases of an intron) are analysed. Other non-coding variants are not currently analysed, unless they were predicted to impact splicing by SpliceAI (Jaganathan et al., 2019) or previously reported in the literature as pathogenic.

On average 97% of coding region exons +-20 base pairs will be covered at greater than 20x, whilst 89% of the entire genome will be covered at a minimum of 20x.

Given typical sequencing conditions the lower confidence interval of the sensitivity of variant calling for Single Nucleotide Variant (SNV) detection is 99.8%. The lower confidence interval of the sensitivity of variant calling for Indel detection is 97.8%. The sensitivity was calculated over a set of high confidence genome regions which are accessible via NGS. The sensitivity of variant calling outside of these regions is unknown.

It is well established that some regions of the genome are inherently difficult to sequence e.g. pseudogene and GC rich regions, and result in low coverage and other issues that may compromise the sensitivity of variant calling. In the event of a negative result, please contact the laboratory to discuss the coverage of any genes that may be considered a likely cause of the patient's phenotypic features.

It is possible that there is a pathogenic variant in the genome which has not currently been detected by this primary analysis. Variants may be identified but not recognised as causative due to insufficient scientific knowledge about the genomic region and its function at the time.

Reanalysis of the data to incorporate updated clinical information and/or newly emerging gene and variant information may be performed and reported at a later time.

Recessive carrier status will not be disclosed.

**** As this test is new to the laboratory's testing repertoire, it has not currently been assessed by UKAS against the ISO15189 standards and is therefore not within the scope of accreditation held by the laboratory****

Samples required

Please send samples in EDTA to the laboratory. We require at least 1ml from the proband and 3-4ml from the parents.

Reporting times

The reporting time for this test is 14 calendar days.

References

1. Kingsmore SF et al. A Randomized, Controlled Trial of the Analytic and Diagnostic Performance of Singleton and Trio, Rapid Genome and Exome Sequencing in Ill Infants. Am J Hum Genet. 2019 Oct 3;105(4):719–33.
2. Mestek-Boukhibar L et al. Rapid Paediatric Sequencing (RaPS): Comprehensive real-life workflow for rapid diagnosis of critically ill children. J Med Genet. 2018.
3. Ceyhan-Birsoy O et al. Interpretation of Genomic Sequencing Results in Healthy and Ill Newborns: Results from the BabySeq Project. Am J Hum Genet. 2019 Jan 3;104(1):76–93.
4. Ceyhan-Birsoy O, Machini K, Lebo MS, Yu TW, Agrawal PB, Parad RB, et al. A curated gene list for reporting results of newborn genomic sequencing. Genet Med. 2017 Jul 1;19(7):809–18.
5. Farnaes L et al. Rapid whole-genome sequencing decreases infant morbidity and cost of hospitalization. npj Genomic Med. 2018 Dec 1;3(1).
6. French CE et al. Whole genome sequencing reveals that genetic conditions are frequent in intensively ill children. Intensive Care Med. 2019 May 1;45(5):627–36.
7. Gubbels CS et al. Prospective, phenotype-driven selection of critically ill neonates for rapid exome sequencing is associated with high diagnostic yield. Genet Med.
8. Holm IA et al. Returning a Genomic Result for an Adult-Onset Condition to the Parents of a Newborn: Insights From the BabySeq Project. Vol. 143, PEDIATRICS. 2019.
9. Petrikin JE et al. The NSIGHT1-randomized controlled trial: Rapid whole-genome sequencing for accelerated etiologic diagnosis in critically ill infants. npj Genomic Med. 2018 Dec 1;3(1).
10. Lindstrand A et al. From cytogenetics to cytogenomics: whole-genome sequencing as a first-line test comprehensively captures the diverse spectrum of disease-causing genetic variation underlying intellectual disability. Genome medicine. 2019 Dec 1;11(1):68.
11. Jaganathan et al. Predicting Splicing from Primary Sequence with Deep Learning. Cell. 2019 Cell, Jan 176(3):414-416