

## ReproXpanded

### Testing for Carrier Status and Adverse Reproductive Outcomes Using Clinical Exome Sequencing

#### Overview:

Genetic changes in many different genes can cause adverse reproductive outcomes, including fetal losses and infertility. Some of these genetic changes may be inherited from both parents and may cause a severe condition in the child, while others may be present in one parent and result in infertility in that individual. Traditional carrier screening has relied heavily on family history and ethnicity information; however, this approach can be limited by inaccurate or incomplete knowledge (Committee Opinion No. 690, 2017; Edwards et al., 2015). Screening for many conditions simultaneously by using high-throughput sequencing can address these limitations and help explain adverse reproductive outcomes. In studies using exome sequencing to detect carrier status, an average of 5-10 deleterious variants were detected per individual screened (Sallevelt et al., 2016; Gambin et al., 2015). Exome sequencing has also been used to identify changes in genes leading to infertility, such as primary ovarian insufficiency and sperm morphology abnormalities, as well as recurrent pregnancy loss and fetal demise (Patiño et al., 2017; Qin et al., 2015; Amiri-Yekta et al., 2016; Ray et al., 2017; Tsuraskaki et al., 2014; Qiao et al., 2016; Yates et al., 2017).

ReproXpanded uses exome sequencing to identify risk to have a child with an autosomal recessive or X-linked condition and to test for genetic changes that can cause adverse pregnancy outcomes. The test uses exome capture, NextGeneration sequencing, and targeted analysis of data to assess known disease genes. This test may be considered for individuals with:

- A previous pregnancy loss or deceased child with no DNA available from the proband
- Unexplained infertility
- Multiple miscarriage of unknown etiology

While expanded carrier screening panels are limited to detecting carrier status for the specific genes or variants included in the panel, ReproXpanded allows for dynamic analysis of all genes that are associated with disease. As carrier status for some types of variants cannot be detected by exome capture, this test is not a replacement for standard carrier screening. ReproXpanded can provide a more comprehensive assessment of reproductive risk when used in combination with standard carrier screening.

## Requirements for ReproXpanded:

- Individuals must be at least 18 years of age
- ReproXpanded must be ordered by a physician
- Relevant clinical and family history information should be submitted with sample(s) to allow for timely and accurate interpretation of genetic data
- ReproXpanded Test Requisition and Informed Consent and Authorization Form

## Result Reporting:

An independent analysis of exome sequence data is performed on each individual submitting a specimen, with a report issued for each person. If samples from both reproductive partners are received at the same time, they will also be analyzed together to assess combined reproductive risk. All results should be discussed with a genetics healthcare provider.

The ReproXpanded report will include:

- Carrier status for pathogenic and likely pathogenic variants in known autosomal recessive and X-linked disease genes
- Carrier status for variants of uncertain significance **only** if the reproductive partner carries a pathogenic or likely pathogenic variant in the same gene
- Pathogenic and likely pathogenic variants in genes associated with infertility or other adverse reproductive outcomes

## Test Methods:

If provided, an individual's clinical records, family history, and prior genetic testing results will be reviewed prior to analysis. Using genomic DNA from the submitted specimen(s), the exonic regions and flanking splice junctions of the genome are captured using a proprietary system developed by GeneDx and sequenced by massively parallel (NextGen) sequencing on an Illumina sequencing system with 100bp or greater paired-end reads. Reads are analyzed for sequence variants using a custom-developed analysis tool (Xome Analyzer). Capillary sequencing or another appropriate method is used to confirm all potentially pathogenic variants identified in the individual. Sequence alterations are reported according to the Human Genome Variation Society (HGVS) nomenclature guidelines. When applicable, phenotype-driven gene lists may be generated using Human Phenotype Ontology (HPO) and Human Gene Mutation Database (HGMD) gene-phenotype associations. Additional resources such as 1000 Genomes database, NHLBI Exome Sequencing Project, ExAC, OMIM, PubMed, and ClinVar are used to evaluate genes and sequence changes of interest, which are then interpreted according to the American College of Medical Genetics and Genomics guidelines (Retterer et al., 2016; Richards et al., 2015).

## Limitations:

Some types of genetic disorders, such as those due to nucleotide repeat expansion/contraction, abnormal DNA methylation, intronic variants, genomic deletions, duplications, insertions or rearrangements, and other mechanisms are not detectable with this test. Specifically, carrier status for spinal muscular atrophy and fragile X syndrome are not identified by this test.

***The ReproXpanded test should therefore not be used as a replacement for standard population-based carrier screening.***

The ReproXpanded test attempts to evaluate the most important regions in the majority of the ~20,000 genes in the human genome. However, it is not technically possible to capture and sequence the entire exome at present. It is anticipated that approximately 95% of the targeted region of an individual's exome will be assessed with the ReproXpanded test at 10x coverage, while >98% of the target region will be covered at a minimum of 1x. There are genes or portions of genes that are not amenable to capture, sequencing, and alignment.

The available scientific knowledge about the function of all genes in the human genome is incomplete at this time. It is possible that ReproXpanded will identify the presence of a genetic variant in the exome sequence of an individual which will not be recognized as causative for a genetic disorder. Reanalysis of the data to incorporate updated clinical information and/or newly emerging gene and variant information is available for a fee. If the ReproXpanded test identifies carrier status in an individual, it is possible that this result will not permit an accurate prediction of the prognosis or severity of the disease in an affected child. While there is a possibility that knowledge of a disorder may help direct management and treatment of a disease, it is also possible that this knowledge will not change management or treatment.

## References:

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