Helix THIS IS A DIAGNOSTIC TEST

Helix Hereditary Multi-Cancer Panel

Patient Name: John Doe	**Patient ID:** 0123456	**Collection Date:** 12-20-2024
Date of Birth: 02-20-1975	**Helix ID:** TST12345	**Order Date:** 12-20-2024
Sex Assigned at Birth: MALE	**Provider Name:** Client Client	**Report Date:** 01-03-2025
Specimen Type: WHOLE BLOOD	**Provider Address:** -	

Results UNCERTAIN

Classification	Gene	DNA Change	Protein Change	Zygosity	Inheritance
UNCERTAIN SIGNIFICANCE	MSH6	c.2882G>C	p.Arg961Thr	Heterozygous	AD

This test did not detect any clinically significant variants within the analyzed gene(s).
Variant(s) of Uncertain Significance were detected in the MSH6 gene.

The clinical impact of the identified variant(s) is uncertain; there is insufficient evidence to determine if there is a disease association. Alteration to medical management is NOT recommended based solely on this result and should instead be guided by personal medical and family history.

The MSH6 gene is associated with the following condition(s):
- autosomal dominant Lynch syndrome (MedGen UID: 318886)
- autosomal recessive constitutional mismatch repair deficiency syndrome (CMMRD) (MedGen UID: 1733656)

Additional clinical information and/or testing biological family members is unlikely to assist in the reclassification of the detected variant(s) at this time.

The Variant Interpretation section below may provide additional details regarding the reported variant(s). Genetic test results should be interpreted in the context of an individual's personal medical and family history. It is important to note that this assay cannot detect all variants known to increase disease risk. Genetic counseling is recommended. Clinical correlation is advised.

Test Description

This panel evaluates 70 genes associated with hereditary cancer conditions that predispose to a variety of primarily adult-onset solid tumors across many organ systems including: breast, gynecologic (ovarian and uterine), colorectal, pancreatic, prostate, kidney, skin, brain and nervous system, and endocrine glands (adrenal, pituitary, parathyroid, thyroid).

Genes Tested

AIP, ALK, APC, ATM, AXIN2, BAP1, BARD1, BLM, BMPR1A, BRCA1, BRCA2, BRIP1, CDC73, CDH1, CDK4, CDKN1B, CDKN2A, CHEK2, CTNNA1, DICER1, EGFR, EPCAM, FH, FLCN, GREM1, HOXB13, KIT, LZTR1, MAX, MBD4, MEN1, MET, MITF, MLH1, MSH2, MSH3, MSH6, MUTYH, NF1, NF2, NTHL1, PALB2, PDGFRA, PMS2, POLD1, POLE, POT1, PRKAR1A, PTCH1, PTEN, RAD51C, RAD51D, RB1, RET, SDHA, SDHAF2, SDHB, SDHC, SDHD, SMAD4, SMARCA4, SMARCB1, SMARCE1, STK11, SUFU, TMEM127, TP53, TSC1, TSC2, VHL

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Transcript:	Genomic Change:
NM_000179.3	NC_000002.12:g.47800865G>C

Variant Interpretation

This variant (NM_000179.3:c.2882G>C p.Arg961Thr) results in the substitution of arginine with threonine at codon 961 in the MSH6 protein. It is present in the gnomAD population database (PMID: 32461654) at the highest allele frequency in the European (non-Finnish) subpopulation (6/1179914 alleles, 0.000509%). This variant has been observed in an individual with with a personal or family history of breast, ovarian, and/or colorectal cancer and the same protein change was observed in an individual with cutaneous melanoma (PMID: 34326862, 29684080). In silico prediction from the HCI Database of Prior Probabilities of Pathogenicity suggests that this variant may be benign. This variant is present in ClinVar (Variation ID: 229829). In conclusion, although there is some evidence suggesting that this variant is benign, additional evidence is necessary to support that, and the clinical role of this variant is unclear at this time. Therefore, it is classified as a Variant of Uncertain Significance.

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Methods & Limitations

Extracted DNA is enriched for targeted regions and then sequenced using the Helix Exome+ (R) assay on an Illumina DNA sequencing system. Data is then aligned to a modified version of GRCh38 and all genes are analyzed using the MANE transcript and MANE Plus Clinical transcript, when available. Small variant calling is completed using a customized version of Sentieon's DNAseq software, augmented by a proprietary small variant caller for difficult variants. Copy number variants (CNVs) are then called using a proprietary bioinformatics pipeline based on depth analysis with a comparison to similarly sequenced samples. Reportable variants in PMS2 exons 12-15 are confirmed by PacBio long reads. The MSH2 Boland inversion (exons 1-7) is detected by identifying discordant read-pairs spanning the presumed breakpoint. Interpretation is based upon guidelines published by the American College of Medical Genetics and Genomics (ACMG) and the Association for Molecular Pathology (AMP) or their modification by ClinGen Variant Curation Expert Panels when available. Interpretation is limited to the transcripts indicated on the report, +/- 10 bp into intronic regions, except as noted below. Helix variant classifications include pathogenic, likely pathogenic, variant of uncertain significance (VUS), likely benign, and benign. Variants classified as pathogenic, likely pathogenic, or VUS are included in the report. All reported variants (except for VUSs with limited evidence of pathogenicity) are confirmed through secondary manual inspection of DNA sequence data or orthogonal testing. Benign and likely benign variants are not reported but are available upon request. Risk estimations and management guidelines included in this report are based on analysis of primary literature and recommendations of applicable professional societies, and should be regarded as approximations.

Based on validation studies, this assay delivers > 99% sensitivity and specificity for single nucleotide variants and insertions and deletions (indels) up to 20 bp. Larger indels and complex variants are also reported but sensitivity may be reduced. Based on validation studies, this assay delivers > 99% sensitivity to multi-exon CNVs and > 90% sensitivity to single-exon CNVs. This test may not detect variants in challenging regions (such as short tandem repeats, homopolymer runs, and segment duplications), sub-exonic CNVs, chromosomal aneuploidy, or variants in the presence of mosaicism. Phasing will be attempted and reported, when possible. Structural rearrangements such as inversions, translocations, and gene conversions are not tested in this assay unless explicitly indicated. Additionally, deep intronic, promoter, and enhancer regions may not be covered. It is important to note that this assay cannot detect all variants known to increase disease risk, and that a negative result does not guarantee that the tested individual does not carry a rare, undetectable variant in genes analyzed. Any potential incidental findings outside of these genes and conditions will not be identified, nor reported. The results of a genetic test may be influenced by various factors, including bone marrow transplantation, blood transfusions, or in rare cases, hematolymphoid neoplasms.

Gene Specific Notes:
APC: analysis includes CNV of promoters 1A and 1B and sequencing of promoter 1B; BMPR1A: analysis includes CNV of promoter; BRCA1: sequencing analysis extends to CDS +/-20 bp; BRCA2: sequencing analysis extends to CDS +/-20 bp. CDKN2A: analysis includes sequencing of the p16 (p16INK4a) and p14 (p14ARF) transcripts; EGFR: analysis is limited to the NM_005228(EGFR):c.2369C>T (p.Thr790Met) variant; EPCAM: analysis is limited to CNV of exons 8-9; GREM1: analysis is limited to CNV of the promoter; HOXB13: analysis is limited to the NM_006361.6(HOXB13):c.251G>A (p.Gly84Glu) variant; MITF: analysis is limited to the NM_000248.4(MITF):c.952G>A (p.Glu318Lys) variant; MLH1: analysis includes CNV of the promoter; MSH2: analysis includes detection of the Boland inversion (inversion of exons 1-7) and detection of NM_000251.3(MSH2):c.942+3A>T; MSH3: analysis excludes sequencing of exon 1 repeat region (chr5:80654878-80654946); POLD1: CNV analysis is not performed and sequencing is limited to the 3'-5' exonuclease domain (chr19:50402681-50407039); POLE: CNV analysis is not performed and sequencing is limited to the 3'-5' exonuclease domain (chr12:132676653-132672296); PTCH1: sensitivity of exon 1 analysis may be reduced; PTEN: analysis includes CNV of the promoter; SDHA: analysis excludes CNV; STK11: sensitivity of exon 3 analysis may be reduced; TP53: analysis includes CNV of the promoter; TSC1: sensitivity of exon 21 analysis may be reduced; VHL: analysis excludes coverage of the cryptic E1' exon (chr3:10142758-10143009)

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Disclaimer

This test was developed and validated by Helix, Inc. This test has not been cleared or approved by the United States Food and Drug Administration (FDA). The Helix laboratory is accredited by the College of American Pathologists (CAP) and certified under the Clinical Laboratory Improvement Amendments (CLIA #: 05D2117342) to perform high-complexity clinical tests. This test is used for clinical purposes. It should not be regarded as investigational or for research.

Reports Signed By

Philip D Cotter, PhD, FACMG, FFSC (RCPA)

Helix's Sequence Once, Query Often ® Model

When your provider first orders a genetic test through Helix, Helix leverages its proprietary Sequence Once, Query Often ® model to perform whole exome sequencing and interpret the specific genes related to the test being ordered. Helix will then continue to store your genetic information for future clinical use. This means that, with your permission, your health care providers can order future medically necessary genetic tests from Helix without the need for you to submit another sample in most cases. Instead, future tests will be performed through digital analysis of your genetic information that is stored by Helix.

When you receive a genetic test performed by Helix, you are in control of how and when your genetic information is used. To manage your genetic information and understand your rights, please visit https://www.helix.com/privacy-and-policy-highlights.