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Variant Report – Comprehensive Solid Tumor Genomic Panel

ORDER DETAILS

Patient

NAME	**HELIX, Onco**
MEDICAL RECORD#	**11111-55555**
DATE OF BIRTH	**July XX, 1918**
GENDER	**Male**
DIAGNOSIS	**Non-small cell lung cancer**

Physician

NAME	**Dr. XXXXX ZZZZZZ**
ORGANIZATION	**S.H.I.E. Logistics Directorate**
ADDRESS	**The Triskelion, Theodore Roosevelt Island**

Specimen and Test

SPECIMEN TYPE	**FFPE**
SPECIMEN ID	**111-222-5555**
TUMOR CELLULARITY (%)	**20**
DATE RECEIVED	**November 15, 2019**
ANALYSIS DATE	**December 5, 2019**
REPORT DATE	**December 13, 2019**
REPORT STATUS	**Verified**

REPORT SUMMARY

Tier 1: Variants of Strong Clinical Significance

VARIANT	LEVEL	VAF%	CLINICAL IMPACT
EGFR p.S768I Missense	**A**	**20**	**Associated with sensitivity to EGFR TKI therapy** - Erlotinib, Gefitinib, Afatinib, Osimertinib, Dacomitinib
KRAS p.G12D Missense	**A**	**18**	**Non-responsive to** - Erlotinib, Gefitinib, Afatinib, Osimertinib, Dacomitinib

Tier 2: Variants of Potential Clinical Significance

VARIANT	LEVEL	VAF%	CLINICAL IMPACT
BRAF p.V600E Missense	**C**	**23**	**Responsive to Dabrafenib,** **Non-responsive to** - Erlotinib, Gefitinib, Afatinib, Osimertinib, Dacomitinib

Gene Amplifications

None

CLINICALLY RELEVANT RESULTS

Tier 1: Variants of Strong Clinical Significance

VARIANT	INTERPRETATION
EGFR p.S768I Missense Level: A	Somatic mutations in the tyrosine kinase domain of the epidermal growth factor receptor (EGFR) gene are present in approximately 80% of the lung adenocarcinomas that respond to EGFR inhibitors (eg, gefitinib, erlotinib and afatinib). EGFR exon19 deletions, exon 21 L858R and Exon 18 mutations correlate strongly with sensitivity to specific EGFR inhibitors, and the response rate to therapy with TKIs has been reported to be up to 80% in such cases. The T790M mutation in exon 20 is associated with resistance to some EGFR inhibitors. However, third generation TKI (eg, osimertinib) can specifically target T790M. Compound (dual) mutations in EGFR have been previously reported in lung adenocarcinoma and typically include a strong activating mutation combined with a weaker activating mutation. These cases appear to respond well to the EGFR targeted therapies. Mutations at E709 in exon 18 often occur together with other mutations in EGFR. This particular complex deletion insertion variant results in both the E709V and G719C in the protein, as well as a K713R variant, which also has been reported previously
KRAS p.G12D Missense Level A	The KRAS protein has intrinsic GTPase activity and is an important mediator of growth factor receptor signaling resulting in the activation of several downstream pathways such as PI3K-mTOR and RAS-RAF-MEK pathway (RefSeq, Jul 2008). A missense alteration in KRAS, G12D, is identified in this case. Codon 12 lies within a GTP binding region of the KRAS protein (UniProt.org). Mutations in KRAS at codon 12 (within the GTP binding region), including KRAS G12D, result in reduced GTPase activity, which in turn leads to constitutive activation of KRAS and its downstream PI3K-AKT and MAPK signaling pathways (PMID-26902995; 25705018). KRAS G12D is reported in malignancies including non-small cell lung cancer (COSMIC, February 2019). Approximately 25% of patients with lung adenocarcinomas in a North American population have KRAS mutations (NCCN, NSCLC v3.2019). KRAS mutation prevalence has been associated with cigarette smoking (NCCN, NSCLC v3.2019). Mutations in KRAS have been associated with reduced responsiveness to EGFR TKI therapy and do not appear to affect chemotherapeutic efficacy (NCCN, NSCLC v3.2019). Targeted therapy is currently not available for patients with KRAS mutations, although immune checkpoint inhibitors appear to be effective; MEK inhibitors are in clinical trials (NCCN, NSCLC v3.2019).

Tier 2: Variants of Potential Clinical Significance

VARIANTS	INTERPRETATION
BRAF p.V600E Missense Level: C	B-RAF is a member of the RAF-family of kinases which plays an important role in the RAS-RAF-MEK-ERK mitotic signaling pathway. Mutations of B-RAF have been described in up to 40-70% of Langerhans cell histiocytosis and approximately 50% of Erdheim-Chester disease. The hotspot for mutations in BRAF is at codon Val600 and these are activating mutations. The most common activating mutation is p.Val600Glu(V600E). Various B-Raf inhibitors(Vemurafenib, Dabrafenib) have been FDA approved for therapy for some tumor types in certain settings, and clinical trials for advanced BRAF V600 mutation-positive tumors using targeted therapy (often in combination with other therapy) may be available (clinical trials.gov).

CLINICAL TRIALS

TITLE	TRIAL IDENTIFIER	PHASE	VARIANT
Bortezomib in KRAS-Mutant Non-Small Cell Lung Cancer in Never Smokers or Those With KRAS G12D	**NCT01833143** https://clinicaltrials.gov/ct2/show/NCT01833143	II	KRAS p.G12D
Study of Regorafenib in Combination With Oral Methotrexate for KRAS Mutated Non-Small Cell Lung Cancer (NSCLC)	**NCT03520842** https://clinicaltrials.gov/show/NCT03520842	II	**KRAS** p.G12D
A Study to Evaluate the Efficacy and Safety of Toripalimab or Placebo Combined With Chemotherapy in Treatment-naive Advanced NSCLC	**NCT03856411** https://clinicaltrials.gov/ct2/show/NCT03856411	III	**EGFR** p.S768I
Study of Selective BRAF Kinase Inhibitor Dabrafenib Monotherapy Twice Daily and in Combination With Dabrafenib Twice Daily and Trametinib Once Daily in Combination Therapy in Subjects With BRAF V600E Mutation Positive Metastatic (Stage IV) Non-small Cell Lung Cancer.	**NCT01336634** https://clinicaltrials.gov/ct2/show/NCT01336634	II	**BRAF** p.V600E
Dabrafenib and Trametinib in Patients With Non-small Cell Lung Cancer Harboring V600E BRAF Mutation	**NCT03543306** https://clinicaltrials.gov/ct2/show/NCT03543306	II	**BRAF** p.V600E

VARIANTS OF UNCERTAIN CLINICAL SIGNIFICANCE

AKT3 NM_001206729.1 c.1345C>T (p.P449S)	**AKT3** NM_001206729.1 c.1348G>A (p.E450K)	**BRCA2** NM_000059.3 c.8951C>A (p.S2984*)	**BRCA2** NM_000059.3 c.8950T>A (p.S2984T)	**BRCA2** p.E2301K NM_000059.3 c.6901G>A
BRCA2 NM_000059.3 c.6888A>G (p.I2296M)	**KRAS** NM_004985.3 c.491G>A (p.R164Q)	**KRAS** NM_004985.3 c.520G>A (p.G174S)	**MSH2** NM_000251.2 c.1698T>A (p.N566K)	**MSH2** NM_000251.2 c.1691C>A (p.T564N)
MSH2 NM_000251.2 c.1688A>C (p.Y563S)	**RB1** NM_000321.2 c.905C>A (p.S302Y)	**RB1** NM_00321.2 c.58G>T (p.W195C)		

VARIANTS CLASSIFICATION AND LEVELS OF EVIDENCE

The variant classification system used in this report is based on joint consensus recommendations of the Association for Molecular Pathology, American Society of Clinical Oncology, and the College of American Pathologists (J Mol Diagn 2017, 19:4-23). Tiers IA, IB, IIC, IID, III and IV describe variant categories of descending clinical significance in the patient. Variants in Tier IV are not reported in accordance with the consensus recommendations.

Tier 1: Variants of Strong Clinical Significance	Tier 2: Variants of Potential Clinical Significance	Tier 3: Variants of Uncertain Clinical Significance	Tier 4: Benign or Likely Benign Variant
Level 'A' Evidence FDA approved therapy Included in professional guidelines **Level 'B' Evidence** Well-powered studies with consensus from experts in the field	**Level 'C' Evidence** FDA approved therapies for different tumor types. Multiple small published studies with some consensus **Level 'D' Evidence** Preclinical trials or a few case reports without consensus	Not observed at a significant allele frequency in the general or specific subpopulation databases, or pan-cancer or tumor-specific variant database No convincing published evidence of cancer association	Observed at a significant allele frequency in the general or specific subpopulation databases No existing published evidence of cancer association

METHODOLOGY

Experimental Methodology

This test uses targeted next-generation sequencing to analyze coding regions of the most inclusive annotated RefSeq transcript for each of the targeted genes. Target enrichment was performed using TruSight Tumor 170 workflow (Illumina). Sequencing of enriched libraries was performed in multiplex on the Illumina NextSeq using the paired-end, 150 base-pair configuration.

Informatics Methodology: Secondary analysis was performed using SOPHiA Genetics pipeline ILL1IC1S3_FFPE v5.5.9. Appropriate coverage (>500x) was confirmed in >95% of regions spanning known hotspots of clinical importance in the targeted genes. Variants passing the quality filters of minimum read depth 500x and variant frequency >5% are reported. ITDs, insertions and deletions >50bp may not be detected by the NGS assay.

In the absence of confirmed somatic status in current databases, this assay cannot distinguish somatic heterozygous from germline variants. A follow-up germline testing may therefore be indicated.

Interpretation of pathogenicity of detected variants is as of current reports and databases.

Sensitivity of the test is 5% for somatic variants detection. Details on low coverage regions can be provided upon request

Test performance characteristics for this laboratory validated test has been determined by the laboratory accredited by College of Physicians and Surgeons of Alberta (CPSA)

DISCLAIMER

This report assumes that the sample received is from the individual noted by the unique identifiers and has not been contaminated with that of another individual prior to receipt at the Hematology Translational Lab. Rare diagnostic errors may result from sample contamination, genotyping errors or sequence polymorphisms in PCR primer binding sites. HGVS classification is provisional. The interpretation is compiled from currently available sources. The content is subject to change and will be adapted from time to time as such sources are updated. Clinical associations described in this report are based on individual SNVs, Indels, fusions and amplifications, and those based on 'combinations of variants' are not provided. Clinical associations based on a "lack of a variant" is not provided. Some drugs identified in the description of variant significance may not be approved by regulatory bodies (including, but not limited to FDA, EMA or NICE) for a particular use or validated for that use. The user is therefore required to independently validate that such drug may be lawfully used in the territory of prescription.

REVIEW AND APPROVAL

Clinical Review:

[Pathologist's Name and Designation]

Report reviewed by

[Laboratory Scientist's name, designation and contact]

Verified and Approved by

[Laboratory Director's name, designation and contact]

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