PATIENT
DISEASE Lung non-small cell lung carcinoma (NOS)
NAME
DATE OF BIRTH
SEX
MEDICAL RECORD #

PHYSICIAN
ORDERING PHYSICIAN
MEDICAL FACILITY
ADDITIONAL RECIPIENT
MEDICAL FACILITY ID
PATHOLOGIST

SPECIMEN
SPECIMEN ID
SPECIMEN TYPE
DATE OF COLLECTION
SPECIMEN RECEIVED

Companion Diagnostic (CDx) Associated Findings

GENOMIC FINDINGS DETECTED	FDA-APPROVED THERAPEUTIC OPTIONS
EGFR L858R	IRESSA® (gefitinib) TAGRISSO® (osimertinib) TARCEVA® (erlotinib)

Other Short Variants Identified

Results reported in this section are not prescriptive or conclusive for labeled use of any specific therapeutic product.
See *professional services* section for information on the alterations listed in this section as well as any additional detected copy number alterations, gene rearrangements, or biomarkers.

OTHER BIOMARKERS WITH POTENTIAL CLINICAL SIGNIFICANCE

DNMT3A R736H #

PIK3CA H1047Q

 # Refer to appendix for limitation statement relating to detection of alterations in ASXL1, ATM, CBL, CHEK2, DNMT3A, JAK2, KMT2D (MLL2), MPL, MYD88, SF3B1, TET2, TP53, and U2AF1.

Please refer to appendix for Explanation of Clinical Significance Classification and for variants of unknown significance (VUS).

ABOUT THE TEST FoundationOne®Liquid CDx is a next generation sequencing (NGS) assay that identifies clinically relevant genomic alterations in circulating cell-free DNA.

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Interpretive content on this page and subsequent pages is provided as a professional service, and is not reviewed or approved by the FDA.

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Biomarker Findings

Blood Tumor Mutational Burden - 8 Muts/Mb
Microsatellite status - Cannot Be Determined
Tumor Fraction - 15%

Genomic Findings

For a complete list of the genes assayed, please refer to the Appendix.

EGFR amplification, L858R
PIK3CA H1047Q
DNMT3A R736H

10 Therapies with Clinical Benefit
0 Therapies with Lack of Response
19 Clinical Trials

BIOMARKER FINDINGS
Blood Tumor Mutational Burden - 8 Muts/Mb
Microsatellite status - Cannot Be Determined
Tumor Fraction - 15%

GENOMIC FINDINGS		VAF %
EGFR -	amplification	-
	L858R	47.5%

10 Trials see p. 14

THERAPY AND CLINICAL TRIAL IMPLICATIONS
No therapies or clinical trials. See Biomarker Findings section
Unable to determine Microsatellite status due to insufficient evidence of genomic instability.
Tumor fraction is an estimate of the percentage of circulating-tumor DNA (ctDNA) present in a cell-free DNA (cfDNA) sample based on observed aneuploid instability.

THERAPIES WITH CLINICAL BENEFIT (IN PATIENT'S TUMOR TYPE)		THERAPIES WITH CLINICAL BENEFIT (IN OTHER TUMOR TYPE)	
Afatinib	1	Cetuximab	2A
Dacomitinib	1	Panitumumab	
Erlotinib	1		
Gefitinib	1		
Osimertinib	1		

☐ *NCCN category*

GENOMIC FINDINGS	VAF %
PIK3CA - H1047Q	2.7%

10 Trials see p. 16

THERAPIES WITH CLINICAL BENEFIT (IN PATIENT'S TUMOR TYPE)	THERAPIES WITH CLINICAL BENEFIT (IN OTHER TUMOR TYPE)
None	Alpelisib
	Everolimus
	Temsirolimus

☐ *NCCN category*

GENOMIC FINDINGS WITH NO REPORTABLE THERAPEUTIC OR CLINICAL TRIALS OPTIONS

For more information regarding biological and clinical significance, including prognostic, diagnostic, germline, and potential chemosensitivity implications, see the Genomic Findings section.

DNMT3A - R736H ..... p. 7

IMPORTANT NOTE Genomic alterations detected may be associated with activity of certain FDA-approved drugs, however, the agents listed in this report may have varied clinical evidence in the patient's tumor type. Neither the therapeutic agents nor the clinical trials identified are ranked in order of potential or predicted efficacy for this patient, nor are they ranked in order of level of evidence for this patient's tumor type. In the appropriate clinical context, germline testing of *APC*, *BRCA1*, *BRCA2*, *BRIP1*, *MEN1*, *MLH1*, *MSH2*, *MSH6*, *MUTYH*, *NF2*, *PALB2*, *PMS2*, *PTEN*, *RAD51C*, *RAD51D*, *RB1*, *RET*, *SDHA*, *SDHB*, *SDHC*, *SDHD*, *SMAD4*, *STK11*, *TGFBR2*, *TP53*, *TSC1*, *TSC2*, *VHL*, and *WT1* is recommended.

Variant Allele Frequency is not applicable for copy number alterations.

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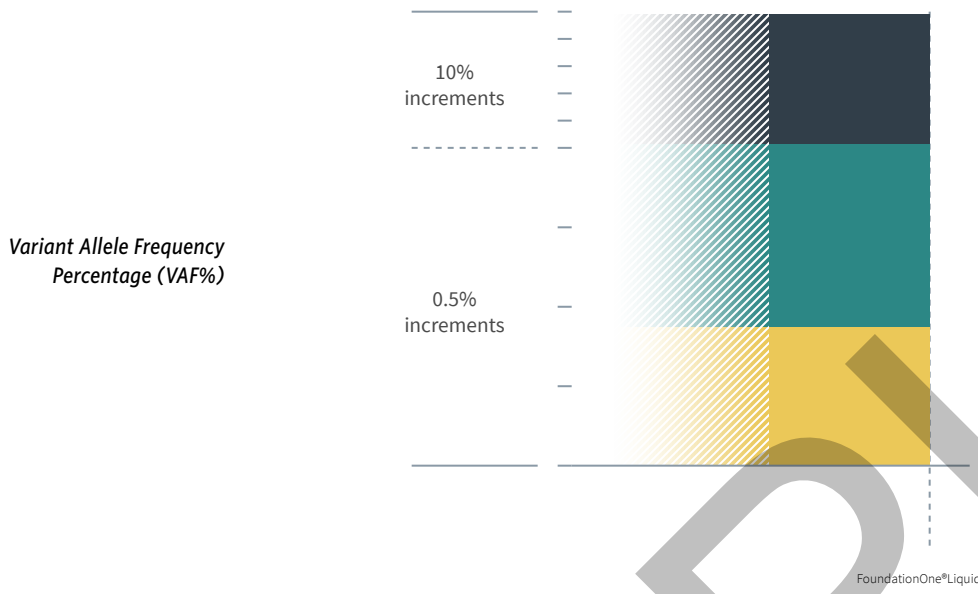

HISTORIC PATIENT FINDINGS		VAF%
Blood Tumor Mutational Burden		8 Muts/Mb
Microsatellite status		Cannot Be Determined
Tumor Fraction		15%
EGFR	● L858R	47.5%
	amplification	Detected
PIK3CA	● H1047Q	2.7%
DNMT3A	● R736H	0.87%

IMPORTANT NOTE This comparison table refers only to genes and biomarkers assayed by prior FoundationOne®Liquid CDx, FoundationOne®Liquid, FoundationOne®, or FoundationOne®CDx tests. Up to five previous tests may be shown.

For some genes in FoundationOne Liquid CDx, only select exons are assayed. Therefore, an alteration found by a previous test may not have been confirmed despite overlapping gene lists. Please refer to the Appendix for the complete list of genes and exons assayed. The gene and biomarker list will be updated periodically to reflect new knowledge about cancer biology.

As new scientific information becomes available, alterations that had previously been listed as Variants of Unknown Significance (VUS) may become reportable.

Tissue Tumor Mutational Burden (TMB) and blood TMB (bTMB) are estimated from the number of synonymous and non-synonymous single-nucleotide variants (SNVs) and insertions and deletions (indels) per area of coding genome sampled, after the removal of known and likely oncogenic driver events and germline SNPs. Tissue TMB is calculated based on variants with an allele frequency of ≥5%, and bTMB is calculated based on variants with an allele frequency of ≥0.5%.

Not Tested = not baited, not reported on test, or test preceded addition of biomarker or gene

Not Detected = baited but not detected on test

Detected = present (VAF% is not applicable)

VAF% = variant allele frequency percentage

Cannot Be Determined = Sample is not of sufficient data quality to confidently determine biomarker status

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BIOMARKER

Blood Tumor Mutational Burden

RESULT
8 Muts/Mb

POTENTIAL TREATMENT STRATEGIES

On the basis of clinical evidence in NSCLC, increased bTMB may be associated with greater sensitivity to immunotherapeutic agents, including anti-PD-L1[1-2] and anti-PD-1[3] therapies. A retrospective analysis of 2 large randomized trials demonstrated patients with NSCLC and a bTMB ≥10 Muts/Mb achieved greater clinical benefit following treatment with atezolizumab than those with bTMB <10 Muts/Mb[1]; similar results have been reported in additional clinical trials using either PD-1 or PD-L1 inhibitors and at higher bTMB cutpoints for patients with NSCLC[3-4] . In a small study, treatment with PD-1 or PD-L1 inhibitors resulted in improved PFS for patients with NSCLC and bTMB ≥6 Muts/Mb as compared to patients with bTMB <6 Muts/Mb[2].

FREQUENCY & PROGNOSIS

NSCLC harbors a median bTMB of 16.8 Muts/Mb (range 1.9-52.5 Muts/Mb)[3]. Published data investigating the prognostic implications of bTMB levels in lung cancer are limited (PubMed, Jul 2020). A large study of Chinese patients with lung adenocarcinoma reported a shorter median OS for tumors with a higher number of mutations in a limited gene set compared with a lower mutation number (48.4 vs. 61.0 months)[5]. Another study of patients with NSCLC correlated elevated TMB with poorer prognosis and significantly associated lower TMB in combination with PD-L1 negative status with longer median survival in patients with lung adenocarcinoma[6]. However, no significant prognostic association of TMB and/or PD-L1 status with survival has been reported in patients with lung SCC[6-7].

FINDING SUMMARY

Blood tumor mutational burden (bTMB, also known as mutation load) is a measure of the number of somatic protein-coding base substitution and insertion/deletion mutations from circulating tumor DNA in blood. TMB is affected by a variety of causes, including exposure to mutagens such as ultraviolet light in melanoma[8-9] and cigarette smoke in lung cancer[10-11], treatment with temozolomide-based chemotherapy in glioma[12-13], mutations in the proofreading domains of DNA polymerases encoded by the POLE and POLD1 genes[14-18], and microsatellite instability (MSI)[14,17-18]. This sample harbors a bTMB below levels that would be predicted to be associated with sensitivity to PD-1- or PD-L1-targeting immune checkpoint inhibitors, alone or in combination with other agents[1-3].

BIOMARKER

Tumor Fraction

RESULT
15%

POTENTIAL TREATMENT STRATEGIES

There are currently no targeted approaches to address specific tumor fraction levels; however, on the basis of emerging clinical evidence, changes in tumor fraction may correlate with treatment duration and clinical response and may be a useful indicator for cancer management[19-24].

FREQUENCY & PROGNOSIS

Detectible ctDNA levels has been reported in a variety of tumor types, with higher tumor fraction levels reported in patients with metastatic (Stage 4) tumors compared with patients with localized disease (Stages 1 to 3)[25]. Elevated tumor fraction levels have been reported to be associated with worse prognosis in a variety of cancer types, including pancreatic cancer[26], Ewing sarcoma and osteosarcoma[27], prostate cancer[22], breast cancer[28], leiomyosarcoma[29], esophageal cancer[30], colorectal cancer[31], and gastrointestinal cancer[32].

FINDING SUMMARY

Tumor fraction is an estimate of the percentage of circulating-tumor DNA (ctDNA) present in a cell-free DNA (cfDNA) sample. Tumor cells in most advanced solid tumor types may shed ctDNA through the process of apoptosis or necrosis[25,33-34]. Tumor fraction has been proposed to be a noninvasive surrogate biomarker of disease burden dynamics. Elevated tumor fraction levels have been associated with inferior prognosis, and therapeutic resistance to treatment in certain tumor types[22,28,31], whereas reduced levels have been correlated with tumor shrinkage and improved clinical outcome in patients with non-small cell lung cancer, urothelial cancer, and melanoma treated with immunotherapy[20,24,35]. The tumor fraction estimate, shown here, is computationally derived from observed aneuploid instability in the sample.

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GENE
EGFR

ALTERATION
amplification, L858R

TRANSCRIPT ID
NM_005228

CODING SEQUENCE EFFECT
2573T>G

POTENTIAL TREATMENT STRATEGIES

EGFR activating mutations may predict sensitivity to EGFR TKIs, including erlotinib[36], gefitinib[37], afatinib[38], dacomitinib[39], and osimertinib[40]. Third-generation EGFR inhibitors, such as osimertinib, selectively target mutated EGFR, including EGFR T790M[40-41]. Osimertinib achieved an ORR of 61% in T790M-positive cases and 21% in T790M-negative cases[40]. Resistance to EGFR inhibition may arise by reactivation of the MAPK pathway, and preclinical evidence suggests that co-targeting EGFR and MAPK signaling may retard the development of acquired resistance to third-generation EGFR inhibitors[42-44]. EGFR amplification or expression may be associated with benefit from anti-EGFR antibodies, such as cetuximab[45-48], panitumumab[46], or necitumumab[49], or EGFR TKIs that target wild-type EGFR[50-54]. Necitumumab is an anti-EGFR antibody that is approved to treat metastatic squamous NSCLC in combination with gemcitabine and cisplatin[55-56] that has also shown benefit in patients with CRC and melanoma[57-58]. Irreversible EGFR inhibitors, as well as HSP90 inhibitors, may be appropriate for patients with de novo or acquired resistance to EGFR-targeted therapy[59-62]. Preclinical studies have reported that EGFR-mutant cells[59-61], including cells with exon 20 insertions[63], are sensitive to HSP90 inhibitors. For patients with EGFR exon 19 deletion/ L858R-positive and T790M- negative NSCLC who had previously progressed on first or second generation EGFR TKIs, a Phase 1 study evaluating the HER3-targeted antibody U3-1402 reported tumor reduction in 12 patients with 2 confirmed PRs (2/13)[64]. Consistent with preclinical data demonstrating that the EGFR inhibitor AZD3759 is capable of penetrating the blood-brain barrier and reducing the volume of brain and leptomeningeal metastases, preliminary results from a Phase 1 trial evaluating single-agent AZD3759 reported a reduction in the volume of brain metastases in 40.0% (8/20) of patients with previously treated NSCLC harboring either EGFR L858R or EGFR exon 19 deletion, including 3 confirmed PRs and 3 unconfirmed PRs[65-66]. In a Phase 1/2 trial for advanced NSCLC, the brain-penetrant third-generation EGFR TKI lazertinib enabled ORRs of 54.3% (69/127) for all evaluable patients and 44.4% (8/18, intracranial) for patients with brain metastases[67]. The reovirus Reolysin targets cells with activated RAS signaling[68-70] and is in clinical trials in patients with some tumor types. Reolysin has demonstrated mixed clinical efficacy, with the highest rate of response reported for patients with head and neck cancer[71-79]. The role of EGFR or KRAS mutations as biomarkers for response to Reolysin in NSCLC is unclear[80]. For patients with NSCLC treated with EGFR tyrosine kinase inhibitors, PIK3CA mutation is associated with shorter OS in a meta-analysis (pooled HR of 1.83)[81]. Clinical studies of lung cancer have shown that acquired PIK3CA mutation may confer resistance to EGFR inhibitors like osimertinib[82-83] . The Phase 3 IMpower study showed that the addition of atezolizumab to bevacizumab plus chemotherapy treatment also had clinical efficacy in patients with untreated EGFR-mutated or ALK-rearranged metastatic NSCLC[84]; therefore, the patient's clinical context should be considered.

FREQUENCY & PROGNOSIS

Amplification of EGFR has been variously reported in 4-42% of non-small cell lung carcinoma (NSCLC) samples[85-89]. EGFR mutation has been reported in 12-36% of lung adenocarcinomas[85,90-91] and in 4% of lung squamous cell carcinomas[86]. EGFR protein expression/overexpression has been reported in up to 70% of NSCLC cases[87-89,92-94]. In addition, expression of EGFR protein has been shown to be higher in lung squamous cell carcinoma samples as compared to lung adenocarcinoma[95-96]. In lung adenocarcinoma, EGFR gene amplification was a predictor of poor disease-free survival in all patients and of poor overall survival in patients with EGFR mutations[97-98]. Nuclear expression of EGFR in NSCLC has been reported to associate with higher disease stage, shorter progression-free survival, and shorter overall survival[99]. However, EGFR mutations have been reported to predict improved survival in patients with resected Stage 1-3 lung adenocarcinoma[100] or resected Stage 1 NSCLC[101].

FINDING SUMMARY

EGFR encodes the epidermal growth factor receptor, which belongs to a class of proteins called receptor tyrosine kinases. In response to signals from the environment, EGFR passes biochemical messages to the cell that stimulate it to grow and divide[102]. Amplification of EGFR has been associated with increased expression of EGFR mRNA and protein in several cancer types[88,103-104]. EGFR L858 is located in the kinase domain and is encoded by exon 21; mutations at this position including L858R[105-107] and L858Q[108] have been characterized as activating. Patients with the L858R mutation have been shown to be sensitive to EGFR tyrosine kinase inhibitors, such as erlotinib, gefitinib[105-107], and afatinib[109]. Other mutations at this position are predicted to be activating.

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