Physician Insert: Oncomine™ Dx Target Test

For In Vitro Diagnostic Use

Epidemiology of Lung Cancer

Lung cancer is the leading cause of cancer death in the United States.[1] In 2017, an estimated 222,500 new cases (116,990 in men and 105,510 in women) of lung and bronchial cancer will be diagnosed, and 155,870 deaths (84,590 in men and 71,280 in women) are estimated to occur because of the disease. [2] Only 17.7% of all patients with lung cancer live 5 years or more after diagnosis. [3]

Genetic Companion Diagnostic Testing for Targeted Therapy Selection in Non-Small Cell Lung Cancer (NSCLC)

Lung cancer is comprised of two main histologic subtypes: non-small cell lung cancer (NSCLC) and small cell lung cancer (SCLC). Over the past decade, several biomarkers associated with therapeutic benefit have emerged for NSCLC. For molecular profiling of other biomarkers, multiplexing technology, such as next-generation sequencing is recommended by IASLC/AMP NSCLC testing guidelines given the limited tissue.

For the most current information on the association of the biomarker and therapeutic outcomes, refer to the therapeutic labels available at Drugs@FDA on the FDA website.

EGFR: EGFR exon 19 deletions and L858R mutation are found in approximately 10% of Caucasian patients with NSCLC and up to 50% of Asian NSCLC patients. [4] These mutations result in activation of the tyrosine kinase domain, and are associated with sensitivity to small molecule tyrosine kinase inhibitors (TKIs), such as erlotinib, gefitinib, and afatinib. [5] Data show that erlotinib, gefitinib, or afatinib (instead of standard first-line chemotherapy) should be used in patients with EGFR exon 19 deletions and L858R mutation.[6 -11] EGFR companion diagnostic tests have been approved by FDA for specific drug indications, including the *therascreen®* EGFR RGQ PCR kit by Qiagen for gefitinib and afatinib, the **cobas®** EGFR Mutation Test v2 by Roche for erlotinib, and the Oncomine™ Dx Target Test by Thermo Fisher Scientific for gefitinib.

ALK: It is estimated that 2-7% of patients with NSCLC have an ALK gene rearrangement.[12] Crizotinib is approved by FDA to treat people with non-small cell lung cancer (NSCLC) that has spread to other parts of the body and is caused by either ALK fusion or ROS1 fusion. Molecular diagnostic testing using FISH and immunohistochemistry (IHC) which are the standard methods for ALK in NSCLC have been approved by FDA for detecting ALK fusion and ALK expression, respectively.[13, 14] While next generation sequencing can also be used to assess presence of ALK fusion, the Oncomine™ Dx Target Test does not detect ALK fusions. To date, two ALK companion diagnostic tests have been approved by FDA for use with crizotinib, including Vysis ALK Break Apart FISH Probe kit by Abbott, and ALK (D5F3) CDx assay by Ventana.

ROS1: It is estimated that ROS1 fusions occur in about 1-2% of patients with NSCLC. [15] ROS1 is very similar to ALK and both are members of the insulin receptor family. Crizotinib is very effective for NSCLC patients with ROS1 rearrangements. [16] The only FDA approved companion diagnostic test which includes detection of the ROS1 fusion is the Oncomine™ Dx Target Test by Thermo Fisher Scientific.

BRAF: It is estimated that BRAF mutations occur in about 2% of patients with NSCLC. [15] Dabrafenib in combination with Trametinib is approved by FDA to treat NSCLC patients with a BRAF V600E mutation. The only FDA approved companion diagnostic test for detection of BRAF V600E in NSCLC is the Oncomine™ Dx Target Test by Thermo Fisher Scientific. There are other FDA approved companion diagnostic tests for BRAF V600E for other indications.

Oncomine™ Dx Target Test

Test Indication

The Oncomine™ Dx Target Test is a qualitative *in vitro* diagnostic test that uses targeted high throughput, parallel-sequencing technology to detect single nucleotide variants (SNVs) and **deletions** in 23 genes from DNA and fusions in ROS1 from RNA isolated from formalin-fixed, paraffin-embedded (FFPE) tumor tissue samples from patients with non-small cell lung cancer (NSCLC) using the Ion PGM™ Dx System.

The test is indicated to aid in selecting NSCLC patients for treatment with the targeted therapies listed in Table 1 in accordance with the approved therapeutic product labeling.

Table 1 - List of variants for therapeutic use

Gene	Variant	Targeted therapy
BRAF	BRAF V600E	TAFINLAR®(dabrafenib) in combination with MEKINIST® (trametinib)
ROS1	ROS1 fusions	XALKORI® (crizotinib)
EGFR	L858R, Exon 19 deletions	IRESSA® (gefitinib)

Safe and effective use of this test has not been established in tissue types other than NSCLC.

Results other than those listed in Table 1 are indicated for use only in patients who have already been considered for all appropriate therapies (including those listed in Table 1).

Analytical performance using NSCLC specimens has been established for the variants listed in Table 2.

Table 2 - List of variants with established analytical performance only

Gene	Variant ID	Nucleotide change
KRAS	COSM512	c.34_35delGGinsTT
KRAS	COSM516	c.34G>T

MET	COSM707	c.3029C>T
PIK3CA	COSM754	c.1035T>A

The test is not indicated to be used for standalone diagnostic purposes, screening, monitoring, risk assessment, or prognosis.

Test Performance and Characteristics

The Oncomine™ Dx Target Test is a qualitative in vitro diagnostic test that uses targeted high throughput, parallel-sequencing technology to detect sequence variations in 23 genes in DNA and RNA isolated from formalin-fixed, paraffin-embedded tumor (FFPE) tissue samples from patients with non-small cell lung cancer (NSCLC) using the Ion PGM™ Dx System.

The 23 genes reported by the test are associated with active NSCLC clinical trials and/or have demonstrated prevalence in literature.

Summary of Reported Variants

Gene	Estimated Prevalence in NSCLC[15,17]	# of Clinical Trials (as of Dec. 2016) [18]
Mutations		
AKT1	0.29%	5
ALK	1.13%	7
BRAF	2.00%	16
CDK4	0.19%	3
DDR2	0.10%	2
EGFR	17.00%	49
ERBB2	2.00%	7
ERBB3	0.00%	2
FGFR2	0.21%	10
FGFR3	0.21%	11
HRAS	0.53%	N/A
KIT	0.00%	3
KRAS	25.00%	13
MAP2K1	0.18%	1
MAP2K2	0.00%	1
MET	3.00%	7
MTOR	0.12%	2
NRAS	0.77%	1
PDGFRA	0.00%	4
PIK3CA	1.34%	8

Gene	Estimated Prevalence in NSCLC[15,17]	# of Clinical Trials (as of Dec. 2016) [18]
RAF1	0.20%	N/A
RET	0.00%	5
ROS1	0.00%	2
Fusions		
ROS1	2.00%	6

Analytical validation of the Oncomine™ Dx Target Test was established through a series of studies to assess the accuracy, sensitivity, specificity, and reproducibility of the assay for the detection of SNVs, deletions and fusions. Based on the data observed with 14 representative variants in 6 genes assessed in clinical samples, the assay demonstrated a limit of detection of 6-13% with 95% confidence for the variant types detected by the test. Additionally, based on 33 representative variants tested across 33 genes, the assay detected the variants with 98.5% positive percent agreement and 100% negative percent agreement against validated comparator detection methods (excluding no calls and invalid results). NPA was calculated as the proportion of reference-negative variant locations for which the Oncomine™ Dx Target Test was also negative. Reproducibility was conducted across four sites; the assay was demonstrated to be reproducible for both positive variant (≥94.5%) and negative variant (≥96.1%) detection across 30 representative variants in 13 **genes** in 18 DNA and 9 RNA samples. For the same representative variants, estimated repeatability was ≥98.8% for DNA and ≥ 94.4% for the ROS1 fusions. These performance studies represent data obtained with the variants assessed. Performance may not be representative of large deletions (>18 bp) or large indels.

The clinical validation of the Oncomine™ Dx Target Test was confirmed for EGFR exon 19 deletions, EGFR exon 21 L858R and BRAF V600E mutations, and ROS1 fusions. Clinical concordance studies showed positive, negative, and overall agreement between the Oncomine™ Dx Target Test results and reference methods.

Guide to Interpreting Results

Test results should be interpreted in the context of pathological evaluation of tumors, treatment history, clinical findings, and other laboratory data.

All clinical interpretations of the variants detected should be made by a board-certified pathologist or equivalent. It is recommended that the physician ordering the test consult with a board-certified pathologist. Patients are advised to seek information from their oncologist or certified health care provider.

Additional information may be obtained from NCCN Guidelines and IASLC/AMP NSCLC Testing Guidelines.

The molecular profile of a tumor can vary between primary and metastatic sites, as well as change over time in response to treatment, leading to the development of mutations that could confer resistance to therapeutic agents.

Test Limitations

The test is designed to interrogate 369 variants in 23 genes. However when certain quality metrics and controls established for the specimen testing are not met, accuracy of the test cannot be assured and therefore mutation status in the exons are reported. Variants detected by the panel, which are not clinically or analytically validated, should not be used for selecting treatment for NSCLC.

This test does not detect genomic copy number variants.

The Oncomine™ Dx Target Test does not detect ALK fusions

This test does not detect structural variants in genes other than ROS1.

Rare polymorphisms exist that could lead to false-negative or false-positive results.

A negative (wild-type) result does not rule out the presence of a mutation that may be present but below the limits of detection of this assay (6-13%).

The product is designed to detect targeted set of known variants in the genes. New variants may be discovered in the future that are not included in the test.

References

1. Torre LA, Siegel RL, Jemal A. Lung cancer statistics. Adv Med Biol 2016;893:1-19
2. Siegel RL, Miller KD, Jemal A. Cancer statistics, 2017. CA Cancer J Clin 2017;67:7-30
3. Howlader N, Noone AM, Krapcho M, et al. SEER Cancer Statistics Review, 1975-2013
4. Hirsch FR, Bunn PA, Jr. EGFR testing in lung cancer is ready for prime time. Lancet Oncol 2009;10:432-433
5. NCCN Guidelines Version 5.2017 Non-Small Cell Lung Cancer
6. Sequist LV, Yang JC, Yamamoto N, et al. Phase III study of afatinib or cisplatin plus pemetrexed in patients with metastatic lung adenocarcinoma with EGFR mutations. J Clin Oncol 2013;31:3327-3334
7. Rosell R, Carcereny E, Gervais R, et al. Erlotinib versus standard chemotherapy as first-line treatment for European patients with advanced EGFR mutation-positive non-small-cell lung cancer (EURTAC): a multicenter, open-label, randomized phase 3 trial. Lancet Oncol 2012;13:239-246
8. Mitsudomi T, Morita S, Yatabe Y, et al. Gefitinib versus cisplatin plus docetaxel in patients with non-small-cell lung cancer harboring mutations of the epidermal growth factor receptor (WJTOG3405): an open label, randomized phase 3 trial. Lancet Oncol 2010;11:121-128
9. Maemondo M, Inoue A, Kobayashi K, et al. Gefitinib or chemotherapy for non-small-cell lung cancer with mutated EGFR. N Engl J Med 2010;362:2380-2388

10. Zhou C, Wu YL, Chen G, et al. Erlotinib versus chemotherapy as first-line treatment for patients with advanced EGFR mutation-positive non-small-cell lung cancer (OPTIMAL, CTONG-0802): a multicenter, open label, randomized, phase 3 study. Lancet Oncol 2011;12:735-742
11. Zhou C, Wu YL, Chen G, et al. Updated efficacy and quality-of-life (QoL) analyses in OPTIMAL, a phase III, randomized, open label study of first-line erlotinib versus gemcitabine/carboplatin in patients with EGFR-activating mutation-positive (EGFR Act Mut+) advanced non-small cell lung cancer (NSCLC). J Clin Oncol 2011;29
12. Kwak EL, Bang YJ, Camidge DR, et al. Anaplastic lymphoma kinase inhibition in non-small-cell lung cancer. N Engl J Med 2010;363:1693-1703
13. Principles of Pathologic Review in the NCCN Guidelines for NSCLC
14. Wynes MW, Sholl LM, Dietel M, et al. An international interpretation study using the ALK IHC antibody D5F3 and a sensitive detection kit demonstrates high concordance between ALK IHC and ALK FISH and between evaluators. J Thorac Oncol 2014;9(5):631-638
15. Tsao AS et al. Scientific Advances in Lung Cancer 2015. J Thorac Onc 2016;11(5):613-638
16. Shaw AT, Ou SH, Bang YJ, et al. Crizotinib in ROS1-rearranged non-small-cell lung cancer. N Engl J Med 2014;371:1963-1971
17. Oncomine_Universal_Dx_Prevalence.20170208, prepared by ThermoFisher Scientific
18. PCP0028358 Attachment 1 Oncomine_Universal_Dx_OKR_annotations, prepared by Thermofisher Scientific