SUMMIT PATHOLOGY
Office located at North Colorado Medical Center
1801 16TH STREET
GREELEY, CO 80631
TEL: (970) 810-6400
FAX: (970) 810-6770

ABNORMAL

SURGICAL PATHOLOGY REPORT

ADDENDED

Patient: GLASTETTER, DENNIS LEON
Med Rec#: **734161** PV: **00102582061**
DOB: **11/16/1955** Age: **64** Sex: **M**
Physician(s):
PAK ALEXANDER NORTH COLORADO MEDICAL CENTER
Attn: **ALEXANDER PAK**

Accession #: 12111507
Date Collected: **06/10/2020**
Date Received: **06/10/2020**
Date Reported: **06/12/2020**
Report Modified: **07/22/2020**

Test Requested: **NCMC Surgical** **Result ID: NS20-02090**

Revision (07/22/2020)
ADDENDUM:

NEO GENOMICS LABORATORIES MOLECULAR GENETICS ANALYSIS REPORT RECEIVED

EGFR Mutation Analysis by Sanger

Results:

Test	Result	Mutations
EGFR Mutation		
EGFR Exon 18	**Not Detected**	**N/A**
EGFR Exon 19	**Not Detected**	**N/A**
EGFR Exon 20 T790M	**Not Detected**	**N/A**
EGFR Exon 20 Other Mutations	**Not Detected**	**N/A**
EGFR Exon 21	**Not Detected**	**N/A**

(See report for details. Report attached) lp

Craig L Nerby, MD
Pathologist, Electronic Signature

Revision (07/20/2020)
ADDENDUM:

NEOGENOMICS LABORATORIES - Molecular Genetics - BRAF Mutation Analysis Report Received

TEST	RESULT
BRAF MUTATION	**NOT DETECTED**

(Please see full report for details. Report is attached.) lr

NEOGENOMICS LABORATORIES - FISH ANALYSIS - ROS1 Report Received

SUMMIT PATHOLOGY
Office located at North Colorado Medical Center
1801 16TH STREET
GREELEY, CO 80631
TEL: (970) 810-6400
FAX: (970) 810-6770

ABNORMAL

SURGICAL PATHOLOGY REPORT

ADDENDED

Patient: GLASTETTER, DENNIS LEON
Med Rec#: **734161** PV: **00102582061**
DOB: **11/16/1955** Age: **64** Sex: **M**
Physician(s):
PAK ALEXANDER
Attn: **ALEXANDER PAK**

NORTH COLORADO MEDICAL CENTER

Accession #: 12111507
Date Collected: **06/10/2020**
Date Received: **06/10/2020**
Date Reported: **06/12/2020**
Report Modified: **07/22/2020**

RESULTS: NEGATIVE

INTERPRETATION:
ROS1 Gene Rearrangement: Not Detected (Negative)

FISH probe signals were within the normal reference range. A ROS1 gene rearrangement was observed in 0% of the nuclei scored and is below the cut-off for this assay. This represents a NEGATIVE result and suggests that ROS1 inhibitors are not indicated.

(Please see full report for details. Report is attached.) lr

NEOGENOMICS LABORATORIES - FISH ANALYSIS - ALK LUNG - Report Received

RESULTS: NEGATIVE

INTERPRETATION:
ALK Rearrangement: Not Detected (Negative)

FISH probe signals were within the normal reference range. An ALK gene rearrangement was observed in 0% of the nuclei scored and is below the threshold of positivity. This represents a NEGATIVE result and suggests that ALK inhibitors are not indicated.

(Please see full report for details. Report is attached.) lr

Craig L Nerby, MD
Pathologist, Electronic Signature

Revision (07/07/2020)
ADDENDUM:

Block tested: A2

PD-L1 (SP263) IHC Results:

SUMMIT PATHOLOGY
Office located at North Colorado Medical Center
1801 16TH STREET
GREELEY, CO 80631
TEL: (970) 810-6400
FAX: (970) 810-6770

ABNORMAL

SURGICAL PATHOLOGY REPORT

ADDENDED

Patient: GLASTETTER, DENNIS LEON
Med Rec#: **734161** PV: **00102582061**
DOB: **11/16/1955** Age: **64** Sex: **M**
Physician(s):
PAK ALEXANDER
Attn: **ALEXANDER PAK**

NORTH COLORADO MEDICAL CENTER

Accession #: 12111507
Date Collected: **06/10/2020**
Date Received: **06/10/2020**
Date Reported: **06/12/2020**
Report Modified: **07/22/2020**

NEGATIVE: <1%
-Tumor proportion score: Negative, less than 1% of cells staining

Dr. Walts has also reviewed this study and concurs.

Interpretive information:

PD-L1 SP263 Ventana Assay performed by immunohistochemistry. Routinely processed formalin-fixed, paraffin-embedded tissues are suitable for use with this primary antibody when used with OptiView DAB IHC detection kit and BenchMark Ultra IHC/ISH instrument. PD-L1 protein expression is determined by using tumor proportion score, which is the presence of viable tumor cells showing partial or complete membrane staining at any intensity above baseline. This assay is indicated as an aid in identifying treatment. Positive and negative controls react satisfactorily.

Arlene Libby, MD
Pathologist, Electronic Signature

Revision (06/25/2020)
ADDENDUM:
MD ANDERSON CANCER CENTER (Annikka Weissferdt, MD) CONSULTATION REPORT RECEIVED

Lung, left lower lobe, wedge resection (A1-A5):
MODERATELY DIFFERENTIATED ADENOCARCINOMA, PRIMARY SITE UNDETERMINED (SEE COMMENT).
TUMOR SIZE: 1.4 CM IN GREATEST DIAMETER (PER REPORT).
LYMPHOVASCULAR INVASION PRESENT.

Lymph node, level 9, biopsy (B1):
One lymph node, no tumor present (0/1).

Lymph nodes, level 10, biopsy (C1-C2):
Two lymph nodes, no tumor present (0/2).

Lymph nodes, level 12, biopsy (D1-D2):
ONE OF THREE LYMPH NODES, POSITIVE FOR PERINODAL LYMPHOVASCULAR INVASION(1/3).

Lymph node, level 5, biopsy (E1):
One lymph node, no tumor present (0/1)

SUMMIT PATHOLOGY
Office located at North Colorado Medical Center
1801 16TH STREET
GREELEY, CO 80631
TEL: (970) 810-6400
FAX: (970) 810-6770

ABNORMAL

SURGICAL PATHOLOGY REPORT

ADDENDED

Patient: GLASTETTER, DENNIS LEON
Med Rec#: **734161** PV: **00102582061**
DOB: **11/16/1955** Age: **64** Sex: **M**
Physician(s):
PAK ALEXANDER NORTH COLORADO MEDICAL CENTER
Attn: **ALEXANDER PAK**

Accession #: 12111507
Date Collected: **06/10/2020**
Date Received: **06/10/2020**
Date Reported: **06/12/2020**
Report Modified: **07/22/2020**

F. Lung, left lower lobe, completion lobectomy (F1-F8):
Lung parenchyma, no residual tumor present.
Bronchial, vascular, and parenchymal margins, free of tumor.
Four peribronchial lymph nodes, no tumor present (0/4).

COMMENT
Submitted immunohistochemical studies preformed on specimen A2 show that the tumor cells are positive for pancytokeratin, CK7, CK5/6 (weak), CK8/18, GATA3 (weak, focal) and negative for TTF1, napsin A, p40, calretinin, CK20, GCDFP-15, MART-1, SOX10, and S100. A submitted special stain for mucicarmine is positive.

Additional immunohistochemical studies performed a MDACC show that the tumor cells are negative for TTF-1, PAX8, and CDX2. Expression of DPV4 (SMAD4) is equivocal.

The morphological and immunohistochemical features are non-specific precluding assessment of the primary site. Based on the predominant tumor arrangement around vascular structures and extensive lymphovascular invasion, a metastatic process should be considered. Close clinical and radiological correlation is advised.

(See report for details. Report scanned, linked and attached.) js

---

Message left for Dr. Pak regarding these results on 6/25/20.

Craig L Nerby, MD
Pathologist, Electronic Signature

FINAL DIAGNOSIS:

A) LUNG, LEFT LOWER LOBE NODULE, WEDGE RESECTION:
1. POSITIVE FOR POORLY DIFFERENTIAED CARCINOMA, **PENDING EXTERNAL CONSULTATION**.
2. SEE COMMENT.

B) LYMPH NODE, LEVEL 9, BIOPSY:
ONE LYMPH NODE, NEGATIVE FOR METASTATIC CARCINOMA (0/1).

C) LYMPH NODES, LEVEL 10, BIOPSY :
TWO LYMPH NODES, NEGATIVE FOR METASTATIC CARCINOMA (0/2).

D) LYMPH NODES, LEVEL12, BIOPSY:
1. ONE OF TWO LYMPH NODES WITH FOCAL PERINODAL LYMPHATIC INVOLVEMENT, SUSPICIOUS

SUMMIT PATHOLOGY
Office located at North Colorado Medical Center
1801 16TH STREET
GREELEY, CO 80631
TEL: (970) 810-6400
FAX: (970) 810-6770

ABNORMAL

SURGICAL PATHOLOGY REPORT

ADDENDED

Patient: GLASTETTER, DENNIS LEON
Med Rec#: **734161** PV: **00102582061**
DOB: **11/16/1955** Age: **64** Sex: **M**
Physician(s):
PAK ALEXANDER NORTH COLORADO MEDICAL CENTER
Attn: **ALEXANDER PAK**

Accession #: 12111507
Date Collected: **06/10/2020**
Date Received: **06/10/2020**
Date Reported: **06/12/2020**
Report Modified: **07/22/2020**

FOR LYMPH NODE METASTASIS (1/2).
2. PENDING EXTERNAL CONSULTATION.

E) LYMPH NODE, LEVEL 5, BIOPSY :
ONE LYMPH NODE, NEGATIVE FOR METASTATIC CARCINOMA (0/1).

F) LUNG, LEFT LOWER LOBE, COMPLETION LOBECTOMY:
1. NO RESIDUAL CARCINOMA IDENTIFIED.
2. FOUR INTRAPARENCHYMAL/PERIBRONCHIAL LYMPH NODES, NEGATIVE FOR METASTATIC CARCINOMA (0/4).
3. MARGINS OF RESECTION ARE NEGATIVE FOR MALIGNANCY.

COMMENT:

The sections in part A demonstrate a carcinoma forming a mass appearing to originate in the perivascular/peribronchial space with foci of lymphovascular invasion identified. The cytologic features are consistent with a poorly-differentiated carcinoma. Immunohistochemical workup demonstrates positivity for pankeratin, CK7, CK5/6, CK8-18, GATA-3, focal napsin-A, and minute focus suggestive of intracellular mucin by mucicarmine stain. The neoplastic cells are negative for CK20, p63, GCDFP-15, S-100, SOX-10, MART-1, TTF-1, calretinin and p40. While the immunohistochemical workup does not entirely exclude the possibility of a lung primary tumor, the panel is not specific for lung primary and the histologic findings are somewhat suggestive of lymphangitic spread/metastatic tumor. Poorly differentiated carcinoma can also lose positivity with some more specific stains. Review of the electronic medical record including a PET scan demonstrates indeterminate, but significant PET avidity in the left parotid gland as well as some PET avidity in the right parotid gland. No prior biopsy of these sites has been performed and thus morphologic comparison is not possible at this time. Slides from this case will be submitted for expert consultation for assistance with classification of the neoplasm. The results of that consultation will be reported in a supplemental report, when available.

Preliminary results discussed with Dr. Pak by phone on 6/12/2020.

Craig L Nerby, MD
Pathologist, Electronic Signature

Clinical Diagnosis: Nodule of left lung

GROSS DESCRIPTION:

A) Received fresh, which has been verified to belong to patient: GLASTETTER, DENNIS LEON and labeled "L lower lobe nodule" as a for g, 4.2 x 2.6 x 2.0 cm lung wedge. The specimen contains a stapled resection margin along the long edge. The pleura is red-tan and smooth, and is inked black. The 0.3 cm in thickness staple line is shaved, and inked orange. Sectioning reveals a 1.4 x 0.8 x 0.8 cm tan-white, firm lesion which abuts orange inked margin, and is 0.2 cm from the pleura. Touch preps are performed, a representative section of the lesion is submitted for frozen section

SUMMIT PATHOLOGY
Office located at North Colorado Medical Center
1801 16TH STREET
GREELEY, CO 80631
TEL: (970) 810-6400
FAX: (970) 810-6770

ABNORMAL

SURGICAL PATHOLOGY REPORT

ADDENDED

Patient: GLASTETTER, DENNIS LEON
Med Rec#: **734161** PV: **00102582061**
DOB: **11/16/1955** Age: **64** Sex: **M**
Physician(s):
PAK ALEXANDER
Attn: **ALEXANDER PAK**

NORTH COLORADO MEDICAL CENTER

Accession #: 12111507
Date Collected: **06/10/2020**
Date Received: **06/10/2020**
Date Reported: **06/12/2020**
Report Modified: **07/22/2020**

evaluation. The remainder of the cut surfaces are red-tan and spongy. The specimen is submitted entirely as follows:
Cassette Summary:
A1: FSA1: Lesion including shaved parenchymal margin
A2: Remainder of lesion and shaved parenchymal margin
A3-A5: Remainder lung

B) Received in a formalin filled bottle/container, which has been verified to belong to patient: GLASTETTER, DENNIS LEON and labeled "level #9 lymph node" is a 0.6 x 0.6 x 0.6 cm gray-tan lymph node with a minimal amount of attached yellow-tan, fatty soft tissue. The specimen is bisected, and submitted entirely in block B1.

C) Received in a formalin filled bottle/container, which has been verified to belong to patient: GLASTETTER, DENNIS LEON and labeled "C level #10 lymph node" are 2 gray-black lymph nodes, 0.6 and 1.2 cm in greatest dimension. The larger lymph node contains a moderate amount of attached yellow-tan soft tissue. The larger lymph node is sectioned. The specimen is submitted entirely as follows:
C1: Larger lymph node
C2: 1 lymph node, intact

D) Received in a formalin filled bottle/container, which has been verified to belong to patient: GLASTETTER, DENNIS LEON and labeled "D level #12 lymph nodes" are 3 gray-black lymph nodes ranging in size from 0.5-1.2 cm. The largest lymph node is trisected. The lymph nodes are submitted entirely as follows:
D1: 1 lymph node, trisected
D2: 2 intact lymph nodes

E) Received in a formalin filled bottle/container, which has been verified to belong to patient: GLASTETTER, DENNIS LEON and labeled "E Level #5 Lymph node" is a 2.0 x 1.6 x 0.6 cm gray-black lymph node with a minimal amount of attached yellow-tan soft tissue. The lymph node is sectioned revealing black cut surfaces. The specimen is submitted entirely in block E1.

F) Received fresh, which has been verified to belong to patient: GLASTETTER, DENNIS LEON and labeled "F L lower lobe" as a 193 g, 19.5 x 11.6 x 4.5 cm left lower lung lobectomy. The bronchial margins are shaved, and submitted for frozen section evaluation. The lung contains 2 stapled parenchymal margins, 7.5 and 10.5 cm in length, the staple lines are shaved, and inked orange. The pleura is purple-pink and smooth with no areas of puckering or induration. Sectioning reveals no definitive masses or lesions. The cut surfaces are red-tan and spongy with diffuse anthracosis. Sectioning reveals 5 gray-black hilar lymph nodes ranging in size from 0.4-1.1 cm.
Representative tissue submitted as follows:
F1: FSF1 - Bronchial margins, en face
F2: Vascular margins, en face
F3-F4: Representative parenchymal margins, perpendicular
F5: Representative lung parenchyma from inferior aspect
F6: 1 lymph node, sectioned
F7: 2 intact lymph nodes
F8: 2 intact lymph nodes

INTRAOPERATIVE CONSULT DIAGNOSIS:

A) Frozen: LLL wedge resection: Non-small cell carcinoma.

Discussed with Dr. Pak on 6/10/20 at 8:54 a.m.

[performed by Catherine Salisbury, MD]

F) Frozen: Bronchial margin:

SUMMIT PATHOLOGY
Office located at North Colorado Medical Center
1801 16TH STREET
GREELEY, CO 80631
TEL: (970) 810-6400
FAX: (970) 810-6770

ABNORMAL

SURGICAL PATHOLOGY REPORT

ADDENDED

Patient: GLASTETTER, DENNIS LEON
Med Rec#: **734161** PV: **00102582061**
DOB: **11/16/1955** Age: **64** Sex: **M**
Physician(s):
PAK ALEXANDER
Attn: **ALEXANDER PAK**

NORTH COLORADO MEDICAL CENTER

Accession #: 12111507
Date Collected: **06/10/2020**
Date Received: **06/10/2020**
Date Reported: **06/12/2020**
Report Modified: **07/22/2020**

Negative for carcinoma.

Discussed with Dr. Pak on 6/10/20 at 10:48 a.m.

[performed by Catherine Salisbury, MD]

MICROSCOPIC DESCRIPTION:

A) One H&E-stained intraoperative touch preparation, one Diff-Quik-stained intraoperative touch preparation, and two H&E-stained slides from one frozen section block are examined. Five H&E slides and 15 immunohistochemical stains (pankeratin, CK7, CK20, CK5/6, CK8-18, p63, p40, TTF-1, GATA-3, GCDFP-15, napsin-A, S-100, SOX-10 and MART-1, and calretinin with appropriate controls) and one histochemical stain (mucicarmine with appropriate control) from five paraffin blocks examined. The immunohistochemical stains were performed to evaluate the neoplastic cells due to the histologic appearance of possible metastatic carcinoma. Please see comment for results of immunohistochemical staining.

B) One H&E slide from one paraffin block examined.

C) Two H&E slides from two paraffin blocks examined.

D) Two H&E slides from two paraffin blocks examined.

E) One H&E slide from one paraffin block examined.

F) Two H&E slides from one frozen section block and eight H&E slides from eight paraffin blocks examined.

NOTE: The immunoperoxidase tests utilized in this examination were developed and their performance characteristics determined by the laboratory at Summit Pathology. It has not been cleared or approved by the U.S. Food and Drug Administration. The FDA has determined that such clearance or approval is not necessary. This test is used for clinical purposes. It should not be regarded as investigational or for research. This laboratory is certified under the Clinical Laboratory Improvement Amendments of 1988 (CLIA) as qualified to perform high complexity clinical laboratory testing.

CPT Code(s): 88313, G9422, 88341 x14, 88360, G9418, 88331 x2, 88305 x4, 88309, 88342, 88307

Specimen grossed and processed at: Summit Pathology 5802 Wright Dr., Loveland, CO, 80538
Specimen interpreted at: North Colorado MC 1801 16th St., Greeley, CO 80631

866.776.5907, option 3

Molecular Genetics

EGFR Mutation Analysis by Sanger

Client 6426
North Colorado Medical Center

1801 16th St
Greeley, CO 80631
Phone: (970) 810-6400
Fax: (970) 810-6770

FX 4

Patient Name: **Glastetter**, **Dennis**
Patient DOB / Sex: **11/16/1955 / M**
Specimen Type: **Paraffin Tissue**
Body Site: **Lung**
Specimen ID: **NS20-02090/NS20-02090-A2**
MRN: **734161**
Reason for Referral: **NODULE OF LEFT LUNG**

Ordering Physician(s): **Craig Nerby, MD**
Treating Physician(s): **Esther Mondo, MD**
Accession / CaseNo: **2909924 / MOL20-225168**
Collection Date: **06/10/2020**
Received Date: **07/14/2020 02:42:00 PM PDT**
Report Date: **07/21/2020 07:16:12 PM EST**

Results:

Test	Result	Mutations
EGFR Mutation		
EGFR Exon 18	Not Detected	N/A
EGFR Exon 19	Not Detected	N/A
EGFR Exon 20 T790M	Not Detected	N/A
EGFR Exon 20 Other Mutations	Not Detected	N/A
EGFR Exon 21	Not Detected	N/A

Clinical Significance:

Patients with non-small cell lung cancer (NSCLC) and mutations in EGFR exons 18, 19, 20 or 21 usually respond to anti-EGFR tyrosine kinase inhibitors (TKIs) and have longer survival when compared to EGFR mutation-negative patients. However, patients being treated with TKIs may develop acquired resistance with secondary mutations in exon 20, such as T790M or, less commonly, L747S, D761Y and T854A.

NSCLCs with exon 20 mutations, such as T790M mutation, are usually resistant to first generation (erlotinib, gefitinib) and second generation (afatinib, dacomitinib, neratinib) anti-EGFR TKIs, but may respond to third generation TKIs, such as osimertinib. The less common resistance mutations (e.g. L747S, D761Y and T854A) may respond to second or third generation irreversible TKIs based on early studies. Most EGFR exon 20 insertions are resistant to EGFR TKIs with the exception of p.A763_Y764insFQEA, which is associated with increased sensitivity to EGFR TKIs.

Methodology:

DNA was isolated from cells or microdissection-enriched FFPE tissue. Formalin-fixed, paraffin-embedded tumor tissue sections were deparaffinized and DNA was isolated. EGFR tyrosine kinase domain mutations were evaluated in the entirety of exons 18 to 21. The patient's sequence is compared to the EGFR sequence database NM_005228. This assay is by Sanger sequencing method with Locked Nucleic Acid (LNA) for T790M. The sensitivity for detecting the T790M mutation in exon 20 is at least 3% with the remaining mutations having a sensitivity of 10 to 15% for detecting mutated EGFR DNA in a wild-type background. Various factors including quantity and quality of nucleic acid, sample preparation and sample age can affect assay performance.

References:

1. Jänne PA, et al. Epidermal growth factor receptor mutations in non-small-cell lung cancer: implications for treatment and tumor biology. J Clin Oncol. 2005; 23:3227-34.
2. Lynch TJ, et al. Activating mutations in the epidermal growth factor receptor underlying responsiveness of non-small-cell lung cancer to gefitinib. N Engl J Med. 2004; 350:2129-39.
3. Sequist LV, et al. Response to treatment and survival of patients with non-small cell lung cancer undergoing somatic EGFR mutation testing. Oncologist. 2007;12:90-8.
4. Paez JG, et al. EGFR mutations in lung cancer: correlation with clinical response to gefitinib therapy. Science. 2004; 304:1497-1500.
5. Costa DB, et al. Differential responses to erlotinib in epidermal growth factor receptor (EGFR)-mutated lung cancers with acquired resistance to gefitinib carrying the L747S and T790M secondary mutations. J Clin Oncol. 2008; 26:1182-4.
6. Felip E, Barlesi F, Besse B, et al. Phase 2 Study of the HSP-90 Inhibitor AUY922 in Previously Treated and Molecularly Defined Patients with Advanced Non-Small Cell Lung Cancer. J Thorac Oncol. 2018;13(4):576-584.

Test/Panel	MolDX CPT	AMA CPT
EGFR Mutation Analysis by Sanger	81235	81235

Electronic Signature

Martin Powers, M.D. Molecular Pathologist - NeoGenomics Lab

The Technical Component Processing, Analysis and Professional Component of this test was completed at NeoGenomics California, 31 Columbia, Aliso Viejo, CA / 92656 / 866-776-5907 / CLIA #05D1021650 / Medical Director(s): Sally Agersborg, M.D.
The performance characteristics of this test have been determined by NeoGenomics Laboratories. This test has not been approved by the FDA. The FDA has determined such clearance or approval is not necessary. This laboratory is CLIA certified to perform high complexity clinical testing.
Images that may be included within this report are representative of the patient but not all testing in its entirety and should not be used to render a result.
The CPT codes provided with our test descriptions are based on MolDX and AMA guidelines and are for informational purposes only. Correct CPT coding is the sole responsibility of the billing party. Please direct any questions regarding coding to the payer being billed.

866.776.5907, option 3

FISH Analysis
ALK Lung

Client 6426
North Colorado Medical Center

1801 16th St
Greeley, CO 80631
Phone: (970) 810-6400
Fax: (970) 810-6770

FX 4

Patient Name: **Glastetter**, **Dennis**
Patient DOB / Sex: **11/16/1955 / M**
Specimen Type: **Paraffin Tissue**
Body Site: **Lung**
Specimen ID: **NS20-02090/NS20-02090-A2**
MRN: **734161**
Reason for Referral: **NODULE OF LEFT LUNG**

Ordering Physician(s): **Craig Nerby, MD**
Treating Physician(s): **Esther Mondo, MD**
Accession / CaseNo: **2909924 / FSG20-063522**
Collection Date: **06/10/2020**
Received Date: **07/14/2020 02:42:00 PM PDT**
Report Date: **07/19/2020 08:24:39 PM EST**

Results: Negative

Interpretation:
ALK Rearrangement: Not Detected (Negative)

FISH probe signals were within the normal reference range. An ALK gene rearrangement was observed in 0% of the nuclei scored and is below the threshold of positivity. This represents a NEGATIVE result and suggests that ALK inhibitors are not indicated.

Methodology: Interphase FISH analysis was performed using the ALK Break Apart FISH Probe Kit. Along with fluorescence in situ hybridization (FISH), an H&E stained slide was reviewed by a pathologist to identify the target area containing invasive tumor. FISH analysis of at least 50 interphase nuclei was performed within the marked target area.

Reference: Pakkala S, Ramalingam SS. Personalized therapy for lung cancer: striking a moving target. JCI Insight. 2018;3(15). PMID: 30089719.

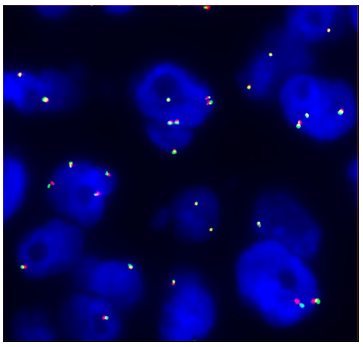
Reference Ranges:
ALK Lung: The sample is considered positive if >50% of the first 50 cells scored are positive, and considered negative if <10% cells are positive. If 10-50% of cells are positive, an additional 50 cells are evaluated by a second technologist. The sample is then considered positive if > or = 15% of all 100 cells scored are positive.

Probe Set Detail:
ALK Lung: nuc ish(ALKx1~>1)[50]

Comments:
The results of this assay have been determined within the limitations described and should not be used interchangeably with resulting values from other methods or kits. These results are intended to be used as an adjunct to other concurrent testing in patient care management. Therefore, the presence or absence of a malignant disease cannot be determined based solely on these results. Clinical correlation is advised.

Nuclei Scored: 50

Probe set	Scoring method	CPT Code	# of Units
ALK Lung	Manual	88377	1



FSG20-063522 Glastetter
Dennis ALK-L ALL.001.A.JPG

Electronic Signature

Saskia Boisot, M.D., Hematopathologist

All controls were within expected ranges.
The Technical Component Processing and Analysis of this test was completed at NeoGenomics California, 31 Columbia, Aliso Viejo, CA / 92656 / 866-776-5907 / CLIA #05D1021650 / Medical Director(s): Sally Agersborg, M.D. The Professional Component of this test was completed at NeoGenomics Rancho Santa Fe, 6463 Paseo Delicias, Rancho Santa Fe, CA / 92067 / 866-776-5907 / Medical Director(s): Saskia Boisot, MD.
NeoGenomics Laboratories FISH test uses either FDA cleared and/or analyte specific reagent (ASR) probes. This test was developed and its performance characteristics determined by the performing laboratory. It has not been cleared or approved by the U.S. Food and Drug Administration (FDA). The FDA has determined that such clearance or approval is not necessary. This test is used for clinical purposes and should not be regarded as investigational or for research. This laboratory is regulated under CLIA '88 as qualified to perform high complexity testing. Interphase FISH does not include examination of the entire chromosomal complement.
Images that may be included within this report are representative of the patient but not all testing in its entirety and should not be used to render a result.
The CPT codes provided with our test descriptions are based on AMA guidelines and are for informational purposes only. Correct CPT coding is the sole responsibility of the billing party. Please direct any questions regarding coding to the payer being billed.

866.776.5907, option 3

Molecular Genetics
BRAF Mutation Analysis

Client 6426
North Colorado Medical Center

1801 16th St
Greeley, CO 80631
Phone: (970) 810-6400
Fax: (970) 810-6770

FX 4

Patient Name: **Glastetter**, **Dennis**
Patient DOB / Sex: **11/16/1955 / M**
Specimen Type: **Paraffin Tissue**
Body Site: **Lung**
Specimen ID: **NS20-02090/NS20-02090-A2**
MRN: **734161**

Ordering Physician(s): **Craig Nerby, MD**
Treating Physician(s): **Esther Mondo, MD**
Accession / CaseNo: **2909924 / MOL20-225170**
Collection Date: **06/10/2020**
Received Date: **07/14/2020 02:42:00 PM PDT**
Report Date: **07/18/2020 05:43:22 PM EST**

Results:

Test	Result
BRAF Mutation	Not Detected

Clinical Significance:

BRAF mutations are frequently found in human cancers. They are found most frequently in melanoma (50-70%), papillary thyroid cancer (36-40%) and most all hairy cell leukemias. BRAF mutations are also found with low frequency in colorectal cancer (5-12%), non-small cell lung cancer (NSCLC), acute myeloid leukemia (AML), glioma, sarcomas, breast cancer, hepatoma, and ovarian cancer. The presence of BRAF mutation is believed to be mutually exclusive to the diagnosis of Lynch syndrome (3).

Patients with BRAF activating mutations, such as those at V600, may respond to therapy including BRAF and MEK inhibitors or anti-VEGF antibodies. BRAF inactivating mutations, such as those at D594, are unlikely to respond to BRAF inhibitors, but may respond to MEK inhibitors. In patients with metastatic colorectal cancer (CRC), the therapeutic significance of BRAF mutation remains controversial. Some studies suggest that it is similar to KRAS mutation, associated with resistance to anti-EGFR therapy. However, recent meta-analysis suggested that patients with BRAF mutation, when treated with anti-EGFR, show similar response to patients without any mutation.

Methodology:

DNA was isolated from cells or microdissection-enriched FFPE tissue. Minimum percentage of tumor in FFPE is at least 20% tumor nuclei out of total nuclei based on pathologist review of an H&E-stained slide. BRAF mutations were evaluated in the entire coding region of BRAF exon 15 by high-sensitivity Sanger sequencing which improves the lower detection limit in mutation hotspot regions to approximately 1% abnormal DNA. This includes V600 mutations and mutations in adjacent codons 598, 599, and 601. Mutation detection outside these hotspot regions has a typical lower detection limit of 10-15% mutated BRAF in a wild-type background. The patient's sequence is compared to the NCBI database: NM_004333. Various factors including quantity and quality of nucleic acid, sample preparation and sample age can affect assay performance.

References:

1. Acquaviva G, Visani M, Repaci A, et al. Molecular pathology of thyroid tumours of follicular cells: a review of genetic alterations and their clinicopathological relevance. Histopathology. 2018;72(1):6-31. PMID: 29239040.
2. Cheng L, Lopez-Beltran A, Massari F, et al. Molecular testing for BRAF mutations to inform melanoma treatment decisions: a move toward precision medicine. Mod Pathol. 2018;31(1):24-38. PMID: 29148538.
3. Chouhan H, Sammour T, Thomas ML, Moore JW. The interaction between BRAF mutation and microsatellite instability (MSI) status in determining survival outcomes after adjuvant 5FU based chemotherapy in stage III colon cancer. J Surg Oncol. 2018;118(8):1311-1317. PMID: 30399198.
4. Jones JC, Renfro LA, Al-Shamsi HO, et al. Non-V600 BRAF Mutations Define a Clinically Distinct Molecular Subtype of Metastatic Colorectal Cancer. J Clin Oncol. 2017;35(23):2624-2630. PMID: 28486044.
5. Karoulia Z, Gavathiotis E, Poulikakos PI. New perspectives for targeting RAF kinase in human cancer. Nat Rev Cancer. 2017;17(11):676-691. PMID: 28984291.
6. Richtig G, Hoeller C, Kashofer K, et al. Beyond the BRAF(V)(600E) hotspot: biology and clinical implications of rare BRAF gene mutations in melanoma patients. Br J Dermatol. 2017;177(4):936-944. PMID: 28278349.
7. Sharma SG, Gulley ML. BRAF mutation testing in colorectal cancer. Arch Pathol Lab Med. 2010;134(8):1225-8. PMID: 20670148.
8. Kreitman RJ. Hairy cell leukemia: present and future directions. Leuk Lymphoma. 2019;60(12):2869-2879. PMID: 31068044.

Test/Panel	MolDX CPT	AMA CPT
BRAF Mutation Analysis	81210	81210

Electronic Signature

Jennifer Hummel, M.D., Pathologist

The Technical Component Processing, Analysis and Professional Component of this test was completed at NeoGenomics California, 31 Columbia, Aliso Viejo, CA / 92656 / 866-776-5907 / CLIA #05D1021650 / Medical Director(s): Sally Agersborg, M.D.
The performance characteristics of this test have been determined by NeoGenomics Laboratories. This test has not been approved by the FDA. The FDA has determined such clearance or approval is not necessary. This laboratory is CLIA certified to perform high complexity clinical testing.
Images that may be included within this report are representative of the patient but not all testing in its entirety and should not be used to render a result.
The CPT codes provided with our test descriptions are based on MolDX and AMA guidelines and are for informational purposes only. Correct CPT coding is the sole responsibility of the billing party. Please direct any questions regarding coding to the payer being billed.

866.776.5907, option 3

FISH Analysis
ROS1

Client 6426
North Colorado Medical Center

1801 16th St
Greeley, CO 80631
Phone: (970) 810-6400
Fax: (970) 810-6770

FX 4

Patient Name: **Glastetter**, **Dennis**
Patient DOB / Sex: **11/16/1955 / M**
Specimen Type: **Paraffin Tissue**
Body Site: **Lung**
Specimen ID: **NS20-02090/NS20-02090-A2**
MRN: **734161**
Reason for Referral: **NODULE OF LEFT LUNG**

Ordering Physician(s): **Craig Nerby, MD**
Treating Physician(s): **Esther Mondo, MD**
Accession / CaseNo: **2909924 / FSG20-063523**
Collection Date: **06/10/2020**
Received Date: **07/14/2020 02:42:00 PM PDT**
Report Date: **07/19/2020 08:25:25 PM EST**

Results: Negative

Interpretation:
ROS1 Gene Rearrangement: Not Detected (Negative)

FISH probe signals were within the normal reference range. A ROS1 gene rearrangement was observed in 0% of the nuclei scored and is below the cut-off for this assay. This represents a NEGATIVE result and suggests that ROS1 inhibitors are not indicated.

Methodology: Interphase FISH analysis was performed using a ROS1 Break Apart FISH Probe. Along with fluorescence in situ hybridization (FISH), an H&E stained slide was reviewed by a pathologist to identify the target area containing invasive tumor. FISH analysis of 50 interphase nuclei was performed within the marked target area.

Reference: Pakkala S, Ramalingam SS. Personalized therapy for lung cancer: striking a moving target. JCI Insight. 2018;3(15). PMID: 30089719.

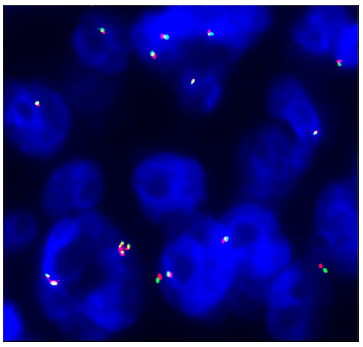
Reference Ranges:
ROS1: The sample is considered positive if >50% of the first 50 cells scored are positive, and considered negative if <10% cells are positive. If 10-50% of cells are positive, an additional 50 cells are evaluated by a second technologist. The sample is then considered positive if > or = 15% of all 100 cells scored are positive.

Probe Set Detail:
ROS1: nuc ish(ROS1x1~>1)[50]

Comments:
The results of this assay have been determined within the limitations described and should not be used interchangeably with resulting values from other methods or kits. These results are intended to be used as an adjunct to other concurrent testing in patient care management. Therefore, the presence or absence of a malignant disease cannot be determined based solely on these results. Clinical correlation is advised.

Nuclei Scored: 50

Probe set	Scoring method	CPT Code	# of Units
ROS1	Manual	88377	1



FSG20-063523 Glastetter
Dennis ROS1 ALL.001.A.JPG

Electronic Signature

Saskia Boisot, M.D., Hematopathologist

All controls were within expected ranges.
The Technical Component Processing and Analysis of this test was completed at NeoGenomics California, 31 Columbia, Aliso Viejo, CA / 92656 / 866-776-5907 / CLIA #05D1021650 / Medical Director(s): Sally Agersborg, M.D. The Professional Component of this test was completed at NeoGenomics Rancho Santa Fe, 6463 Paseo Delicias, Rancho Santa Fe, CA / 92067 / 866-776-5907 / Medical Director(s): Saskia Boisot, MD.
NeoGenomics Laboratories FISH test uses either FDA cleared and/or analyte specific reagent (ASR) probes. This test was developed and its performance characteristics determined by NeoGenomics Laboratories in Aliso Viejo, CA. It has not been cleared or approved by the U.S. Food and Drug Administration (FDA). The FDA has determined that such clearance or approval is not necessary. This test is used for clinical purposes and should not be regarded as investigational or for research. This laboratory is regulated under CLIA '88 as qualified to perform high complexity testing.Interphase FISH does not include examination of the entire chromosomal complement. Clinically significant anomalies detectable by routine banded cytogenetic analysis may still be present. Consider reflex banded cytogenetic analysis. Images that may be included within this report are representative of the patient but not all testing in its entirety and should not be used to render a result.
The CPT codes provided with our test descriptions are based on AMA guidelines and are for informational purposes only. Correct CPT coding is the sole responsibility of the billing party. Please direct any questions regarding coding to the payer being billed.

NS20-02090 REQ
GLASTETTER, DEN
DOB: 11/16/1955
DOS: 06/10/2020
12111507

THE UNIVERSITY OF TEXAS
MD Anderson Cancer Center
1515 HOLCOMBE BOULEVARD
HOUSTON, TX 77030-4095

Consultation Report

Department of Pathology, Box 85
Tel: 713-792-3205 Fax: 713-794-4630

2502061

GLASTETTER, DENNIS

DOB: 11/16/1955 Sex: M
Physician: Craig L Nerby, MD

Accession date: 06/18/2020 09:49

Accession: **S-20-033101**

Case type: Outside Consultation

Mail to: **Summit Pathology**
Pathology
5802 Wright Drive
Loveland CO 80538

MATERIALS RECEIVED

Accession #	Collected	Received	Slides	Blocks	UnstSlides
NS20-02090	06/10/2020	06/17/2020	39	1	0

DIAGNOSIS

Thirty-nine slides and one paraffin block (NS20-02090) designated as follows:

Lung, left lower lobe, wedge resection (A1-A5):

MODERATELY DIFFERENTIATED ADENOCARCINOMA, PRIMARY SITE UNDETERMINED (SEE COMMENT).
TUMOR SIZE: 1.4 CM IN GREATEST DIAMETER (PER REPORT).
LYMPHOVASCULAR INVASION PRESENT.

Lymph node, level 9, biopsy (B1):

One lymph node, no tumor present (0/1).

Lymph nodes, level 10, biopsy (C1-C2):

Two lymph nodes, no tumor present (0/2).

Lymph nodes, level 12, biopsy (D1-D2):

ONE OF THREE LYMPH NODES, POSITIVE FOR PERINODAL LYMPHOVASCULAR INVASION (1/3).

Lymph node, level 5, biopsy (E1):

One lymph node, no tumor present (0/1).

F. Lung, left lower lobe, completion lobectomy (F1-F8):

Lung parenchyma, no residual tumor present.
Bronchial, vascular, and parenchymal margins, free of tumor.
Four peribronchial lymph nodes, no tumor present (0/4).

Consultation Report
File under: Pathology

NS20-02090 REQ
GLASTETTER, DEN
DOB: 11/16/1955
DOS: 06/18/2020
12111507

THE UNIVERSITY OF TEXAS
MD Anderson Cancer Center
1515 HOLCOMBE BOULEVARD
HOUSTON, TX 77030-4095

Consultation Report

Department of Pathology, Box 85
Tel: 713-792-3205 Fax: 713-794-4630

2502061

GLASTETTER, DENNIS
DOB: 11/16/1955 Age: 64 Sex: M
Physician: Craig L Nerby, MD

Received: 06/18/2020 09:49

Accession: **S-20-033101**

Case type: Outside Consultation

COMMENT

Submitted immunohistochemical studies performed on specimen A2 show that the tumor cells are positive for pancytokeratin, CK7, CK5/6 (weak), CK8/18, GATA3 (weak, focal) and negative for TTF1, napsin A, p40, p63, calretinin, CK20, GCDFP-15, MART-1, SOX10, and S100. A submitted special stain for mucicarmine is positive.

Additional immunohistochemical studies performed at MDACC show that the tumor cells are negative for TTF-1, PAX8, and CDX2. Expression of DPC4 (SMAD4) is equivocal.

The morphological and immunohistochemical features are non-specific precluding assessment of the primary site. Based on the predominant tumor arrangement around vascular structures and extensive lymphovascular invasion, a metastatic process should be considered. Close clinical and radiological correlation is advised.

AW/AK
6/21/2020 11:52 AM

BIOMARKER TESTING

Tumor Block: A2

SNOMED CODES

T-28000, M-81406

"Some tests reported here may have been developed and performance characteristics determined by UT MD Anderson Pathology and Laboratory Medicine. These tests have not been specifically cleared or approved by the U.S. Food and Drug Administration. If applicable, controls were reviewed and showed appropriate reactivity."

Final Diagnosis completed by Annikka Weissferdt, MD. Electronically signed report date 6/23/2020 10:28AM