Sample: 16K1023-1 Name: Stott,Stanley DOB: 31-May-1936 URN: 16/11/67320

LUNG CANCER MUTATION ANALYSIS DRAFT

SPECIMEN
extref

PATHOLOGY
Histological typing: .....
The sample was reviewed by a pathologist and was considered to have ....% tumour cells within the area selected for analysis. Please note: This is not a formal pathology review and is based solely on an H&E of the tissue provided and not on ancillary clinical or pathology information that may be available elsewhere.

RESULT

Gene	EGFR
Reference	NM_005228.3
cDNA Change	c.2126A>C
Protein Change	NP_005219.2:p.(Glu709Ala)
Read Depth	2037/7780 26.2%

RESULT

Gene	EGFR
Reference	NM_005228.3
cDNA Change	c.2573T>G
Protein Change	NP_005219.2:p.(Leu858Arg)
Read Depth	2362/10601 22.3%

TEST DESCRIPTION
Tumour DNA was tested in duplicate for mutations in exons 19 to 21 of the EGFR gene, exons 2 to 4 of the KRAS gene, and exon 15 of the BRAF gene using massively parallel sequencing. This test detects single nucleotide variants and indels in the target exons only. At 1000x coverage, the limit of detection of this assay has been determined to be X%. At 500x coverage the limit of detection has been determined to be X%. The sample was sequenced to an average 2504 aligned reads per amplicon with 99.28% uniformity. Regions with less than 100x coverage have not been analysed. These are listed below.

INTERPRETATION
EGFR: CurVariant chr7:g.55241678A>C not yet curated.

EGFR: The L858R mutation results in an amino acid substitution at position 858 in EGFR, from a leucine (L) to an arginine (R). This mutation occurs within exon 21, which encodes part of the kinase domain, and occurs with a frequency of approximately 43% in EGFR mutant lung tumors. In the metastatic setting, EGFR mutations are strong predictors of efficacy for the EGFR tyrosine kinase inhibitors (TKIs), including the "first-generation" drugs erlotinib (Tarceva) and gefitinib (Iressa).

COMMENTS
Mutations in the kinase domain of the epidermal growth factor receptor (EGFR) gene result in constitutive signalling leading to tumour development. Kinase domain mutations occur in approximately 10% of non-South East Asian and 35% of South East Asian NSCLC patients, the majority of which display a dramatic response to EGFR kinase domain inhibitors (1). Confirmation of EGFR mutation status is required before administering kinase domain inhibitors such as gefitinib (Iressa) and erlotinib (Tarceva).
Activating KRAS mutations occur in up to 40% of NSCLC and occur most frequently at codons 12, 13 and 61. KRAS mutations cause constitutive activation resulting in a continual proliferative signal downstream of EGFR. KRAS mutant NSCLC is insensitive to targeted EGFR inhibitors (2). In a retrospective series of 1,046 NSCLC patients, the BRAF V600E mutation was associated with shorter disease free survival (3).

Note: Testing of tissue treated with chemo and/or radiotherapy reduces the cellularity of the neoplastic element and reduces the sensitivity of the assay. Where possible

tissue derived from untreated tumour should be tested.

REFERENCES

1. Mok, T.S., et al., N Engl J Med, 2009. 361(10): p. 947-57.
2. Sun, J.M., et al., PLoS One, 2013. 8(5): p. e64816.
3. Marchetti, A., et al., Journal of Clinical Oncology, 2011. 29(26): p. 3574-3579.

Low coverage amplicons:
There were 0 low read amplicons with <100 aligned reads:
not listed

Assay region of interest coverage:
BRAF_Ex11_444 (coverage 3453)
BRAF_Ex11_460-470 (coverage 3453)
BRAF_Ex15_594-603 (coverage 10857)
EGFR_Ex18_705-720 (coverage 7838)
EGFR_Ex19_Whole-Exon (coverage 6180)
EGFR_Ex20_Whole-Exon (coverage 2350)
EGFR_Ex21_855-865 (coverage 5338)
KRAS_Ex2_12-13 (coverage 5778)
KRAS_Ex3_59-61 (coverage 3362)
KRAS_Ex4_117 (coverage 7070)
KRAS_Ex4_146 (coverage 4705)
MET_Ex14_3-Splice (coverage 3285)