Sample: 14M8152 Name: FOSTER,TRACY DOB: 01-Apr-1966 URN: 14/11/68177

MELANOMA 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	NRAS
Reference	c.182A>T
cDNA Change	c.182A>T
Protein Change	p.Gln61Leu
Read Depth	3872/9872 39.2%

TEST DESCRIPTION
Tumour DNA was tested in duplicate for mutations in exons 11 and 15 of the BRAF gene, exons 11, 13, 17 and 18 of the c-KIT gene, and exons 2 to 4 of the NRAS 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 2877 aligned reads per amplicon with 90.71% uniformity. Regions with less than 100x coverage have not been analysed. These are listed below.

INTERPRETATION
NRAS: The NRAS p.Gln61Leu mutation predicts a missense amino acid substitution at position 61 in GTPase NRas, from a glutamine (Gln, Q) to a leucine (Leu, L). Codon 61 is the predominant mutation hotspot in NRAS with multiple missense variants at this residue resulting in constitutive activation of NRAS signaling pathways. Based on current scientific knowledge, this is a pathogenic variant. The role of NRAS mutations for selecting/prioritising anticancer treatment, including cytotoxic chemotherapy and targeted agents, is unknown at this time however, NRAS mutations have been shown to confer sensitivity to MEK inhibitors (reported in melanoma and preclinically in non-small cell lung carcinoma, [1, 2]). (From mycancergenome.org)

1. Ascierto, P.A., et al., MEK162 for patients with advanced melanoma harbouring NRAS or Val600 BRAF mutations: a non-randomised, open-label phase 2 study. Lancet Oncol, 2013. 14(3): p. 249-56.
2. Ohashi, K., et al., Characteristics of lung cancers harboring NRAS mutations. Clin Cancer Res, 2013. 19(9): p. 2584-91.

COMMENTS
The BRAF V600E mutation occurs in 40 to 60% of malignant melanomas and confers sensitivity to the targeted small molecule inhibitor vemurafinib (1). The NRAS gene is mutated in 20% of malignant melanomas and may be amenable to treatment with downstream MAPK and PI3K inhibitors (2). The KIT gene is mutated in 17% of cutaneous melanomas, 11% of acral melanomas and 21% of mucosal melanomas. Patients with KIT mutations benefit from imatinib therapy (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. Flaherty, K.T., et al. New Engl J Med, 2010. 363(9): p. 809-819.
2. Kelleher, F.C., McArthur, G.A., Cancer J., 2012. 18(2): p. 132-136.
3. Hodi, F.S., et al., Journal of Clinical Oncology, 2013. 31(26): p. 3182-3190.

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

Assay region of interest coverage:
NRAS_Ex2_12-13 (coverage 4535)
NRAS_Ex3_59-61 (coverage 9888)
NRAS_Ex4_117 (coverage 12602)
NRAS_Ex4_146 (coverage 12602)
BRAF_Ex11_460-470 (coverage 3441)
BRAF_Ex15_594-603 (coverage 9998)
KIT_Ex9_500-505 (coverage 3143)
KIT_Ex11_Whole-Exon (coverage 4629)
KIT_Ex13_Whole-Exon (coverage 4125)
KIT_Ex17_Whole-Exon (coverage 3232)