Weill Cornell Medicine
Clinical Genomics Lab

NewYork-Presbyterian

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GX18-xyx

Oncomine Comprehensive Report

Patient Name:	Patient, Sample	**Ordering Physician:**	Philip E. Stieg, M.D.
DOB:	99/99/1999	**Neoplastic content:**	80%
Sex:	M	**Specimen ID:**	S18-XYZ
MRN:	10001001	**Sample Type:**	FFPE
Tumor Type:	Glioblastoma	**Sample Collected:**	8/22/2018
Primary site:	Brain	**Sample Received:**	8/24/2018
Tissue Tested:	Brain	**Date of Service:**	8/22/2018

Tier 1 Variants

Variant	Type	VAF	CN
EGFR	Amplification	N/A	44.96

Tier 2 Variants

Variant	Type	VAF	CN
PIK3R1 c.1043_1049delGAGATAC, p.Arg348fs	frameshiftDeletion	29.5%	N/A
EGFR c.323G>A, p.Arg108Lys	missense	29.4%	N/A
PTEN c.1113delC, p.Asp371fs	frameshiftDeletion	50.8%	N/A
EGFR(1) - EGFR(8)	EGFR vIII	N/A	N/A

Tier 3 Variants

Variant	Type	VAF	CN
EGFR c.680C>T, p.Ser227Phe	missense	92.5%	N/A

VAF = Variant Allele Frequency, CN = Copy Number, N/A = Not Applicable

Comments

EGFR mutations in GBM cluster in the extracellular (EC) domain and include in-frame deletions (such as the common “variant III” del 6-273) and missense mutations (A289V, A289D, T263P, G598V). Mutations involving residue R108 have been reported in GBM, frequently occurring with other EGFR mutations at amino acids A289, P596, and G598. In vitro studies have shown that R108K mutation leads to increased ligand-binding affinity and shows anchorage-independent growth and tumorigenic potential when stably expressed in NIH-3T3 cells. The predictive and prognostic significance of this mutation at R108 needs further elucidation. Correlation with other clinical and laboratory findings is recommended.

Greater than 40% of glioblastomas (GBM) harbor focal amplification of the EGFR locus and there is evidence to suggest that these are driver alterations in these patients, making the EGFR pathway a potential therapeutic target in some clinical settings. Moreover, this alteration is relatively specific for GBM with very few other diffusely infiltrative gliomas having been shown to carry focal amplification of this locus (<3%). In GBM, this alteration frequently occurs in combination with other alterations of EGFR including polysomy 7, intragenic inframe deletions (e.g. EGFRvIII), and/or

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somatic point mutations. Based on current evidence, the independent predictive value of EGFR amplification in GBM is unclear. The relationship between individual and concurrent EGFR alterations and clinical response to small molecular inhibitors targeting EGFR is currently under investigation in clinical trials.

Classification of variants: Variants are classified based on current evidence for clinical actionability.

Tier 1 - Clinical utility has been demonstrated - Actionable / Clinically Relevant variants. Variants in genes with approved therapeutic implications in specified tumors.

Tier 2 - Clinical utility/actionability has diagnostic, prognostic or therapeutic implications. Variants with potential diagnostic/classification, prognostic implications. Variants with approved therapeutic implications in a different tumor type. Novel variants in genes that have approved therapeutic implications. Variants associated with Clinical trials.

Tier 3 - Variants of Unknown Significance.

Method: DNA and RNA were extracted from macrodissected, paraffin-embedded tumor of the patient using the Maxwell 16 instrument (Promega, Madison, WI) and RecoverAll Total Nucleic Acid Isolation Kit (Life Technologies, Waltham, MA), respectively. The extracted DNA and synthesized cDNA from the extracted RNA were amplified by the Oncomine Comprehensive Panel (OCP) and subjected to Next generation Sequencing (NGS) using the Ion Torrent S5™(Life Technologies). The targeted gene panel interrogates 143 unique cancer genes including 73 oncogenes, 49 copy number alteration (CNA) genes, 26 tumor suppressor genes, and 23 fusion driver genes. OCP is designed to detect mutations/single nucleotide variants (SNVs), insertion/deletion (Indel), copy number variants (CNVs), and gene fusions. This test is validated for SNVs in the BRAF, EGFR, KRAS, NRAS, IDH1, and PIK3CA genes, indel in the EGFR gene, CNVs in the HER2 gene, and gene fusions in the ALK and ERG genes. The limit of detection for SNVs and indels is precise and reproducible at 5% with 400X coverage and 3% with 1000X coverage. The minimum number of reads required for fusion positivity is 1000. Genes with 5% CI estimate of ≥4 is considered to have copy number gains. The data obtained was analyzed with the Ion Reporter™ Software 5.6 including Coverage Analysis, Fusion Analysis, and Torrent Variant Annotator v2.3 plug-ins. DNA sequences used as references for this panel of genes can be found at http://ncbi.nlm.nih.gov/refseq/rsg. The mutation nomenclature is based on the recommendations from the Human Genome Variation Society http://www.hgvs.org/mutnomen.

Technical Assessment: This panel is designed to detect mutations in 99 genes, CNVs in 75 genes, and fusions in 23 genes. Out of 130 DNA-sequenced genes, 104 genes are not sequenced in their entirely. Mutations outside the 2530 interrogated amplicons will not be detected. Due to the technology employed by this NGS assay, accurate indel identification in homopolymeric region is not optimal. For 23 fusion genes, 148 isoforms using 154 primer pairs are targeted, but potential isoforms which are not covered by the primer pairs will not be detected.

Disclaimer: The Oncomine Comprehensive Test was developed and its performance characteristics was determined by the Clinical Genomics Laboratory, Englander Institute for Precision Medicine/Department of Pathology and Laboratory Medicine at Weill Cornell Medicine/New York-Presbyterian Hospital; and approved by the New York-State Department of Health (NYS-DOH). This method has not been cleared by the Food and Drug Administration (FDA). The FDA has determined that such clearance or approval is not necessary. Variants of uncertain origin (germline versus somatic origin) cannot be determined unequivocally in this test, such that germline alterations are not reported. If a possible pathogenic germline mutation (inherited) is suspected, then counselling by a board certified genetic counselor will be recommended in note.

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HOTSPOT			FULL GENE	CNV		FUSION
ABL1	GNA11	MYD88	APC	ACVRL1	IL6	ABL1
AKT1	GNAQ	NFE2L2	ATM	AKT1	KIT	AKT3
ALK	GNAS	NPM1	BAP1	APEX1	KRAS	ALK
AR	HNF1A	NRAS	BRCA1	AR	MCL1	AXL
ARAF	HRAS	PAX5	BRCA2	ATP11B	MDM2	BRAF
BRAF	IDH1	PDGFRA	CDH1	BCL2L1	MDM4	ERG
BTK	IDH2	PIK3CA	CDKN2A	BCL9	MET	ETV1
CBL	IFITM1	PPP2R1A	FBXW7	BIRC2	MYC	ETV4
CDK4	IFITM3	PTPN11	GATA3	BIRC3	MYCL	ETV5
CHEK2	JAK1	RAC1	MSH2	CCND1	MYCN	EGFR
CSF1R	JAK2	RAF1	NF1	CCNE1	MYO18A	ERBB2
CTNNB1	JAK3	RET	NF2	CD274	NKX2-1	FGFR1
DDR2	KDR	RHEB	NOTCH1	CD44	NKX2-8	FGFR2
DNMT3A	KIT	RHOA	PIK3R1	CDK4	PDCD1LG2	FGFR3
EGFR	KNSTRN	SF3B1	PTCH1	CDK6	PDGFRA	MET
ERBB2	KRAS	SMO	PTEN	CSNK2A1	PIK3CA	NTRK1
ERBB3	MAGOH	SPOP	RB1	DCUN1D1	PNP	NTRK2
ERBB4	MAP2K1	SRC	SMAD4	EGFR	PPARG	NTRK3
ESR1	MAP2K2	STAT3	SMARCB1	ERBB2	SMARCB1	PDGFRA
EZH2	MAPK1	U2AF1	STK11	FGFR1	SOX2	PPARG
FGFR1	MAX	XPO1	TET2	FGFR2	TERT	RAF1
FGFR2	MED12		TP53	FGFR3	TIAF1	RET
FGFR3	MET		TSC1	FGFR4	ZNF217	ROS1
FLT3	MLH1		TSC2	FLT3		
FOXL2	MPL		VHL	GAS6		
GATA2	MTOR		WT1	IGF1R		

References

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