Brigham and Women's Hospital
Department of Pathology
Center for Advanced Molecular Diagnostics
75 Francis Street, Boston, MA 02115
Tel (857) 307-1540 Fax (857) 307-1544

Accession No.:
Unit Number(s):
Patient Name:
Birth Date:
Age & Sex at Diagnosis:

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Test Performed - MDONPANEL_B

Test Description - OncoPanel

Accession numbers on blocks/tissue submitted – TM-8251980

Original specimen collection date – 11/02/2014

Original pathologic diagnosis – Lung adenocarcinoma

Estimated percentage of neoplastic cells in submitted specimen - 15%

RESULTS:

There are 11588951 aligned, high-quality reads for this specimen with a mean of 298 reads across all targeted exons and 98% of all exons having more than 30 reads.

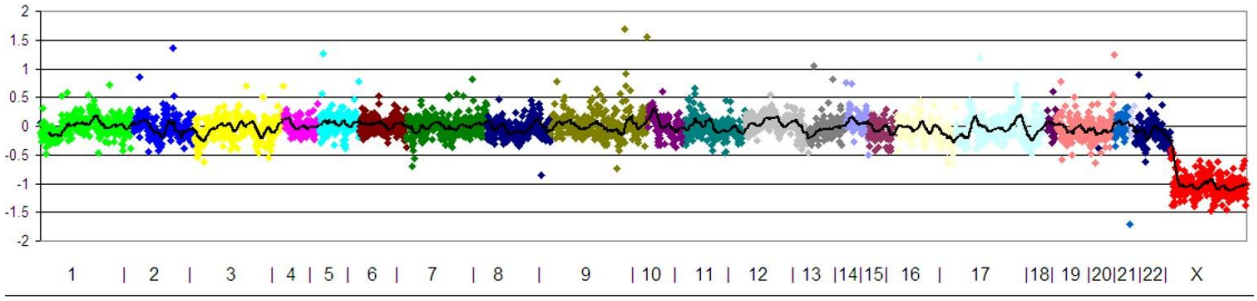

Figure legend: Plot of copy number variation by chromosomes which are color-coded. Sex chromosomes are excluded from the analysis. The vertical axis is the ratio of number of reads for this specimen and a panel of normals in log base 2 scale. A value of 0 denotes no difference from normal (diploid). When the sample contains 100% tumor cells, a value of -1 equals to 1 copy loss and 0.58 is 1 copy gain. The sensitivity and specificity of copy number variation evaluation by next-generation sequencing is affected by several factors, including the tumor percentage, ploidy, clonal heterogeneity, and the GC content of the gene of interest. For example, a sample with 20% tumor cells having a 5-copy amplification of a gene is indistinguishable from a sample with 100% tumor cells with 1 copy gain of the same gene. Confirmation of the copy number variation findings by Next-Gen Sequencing with a different testing platform is recommended.

DNA VARIANTS:

See Background section for tier definitions

Tier 1 variants:

NEGATIVE for mutations in the following genes with clinical relevance for this tumor type: BRAF, EGFR, KRAS, MET

Tier 2 variants: None identified.

CLIA certificate: 22D2040971

Laboratory Director
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Tier 3 variants: None identified.

Tier 4 variants:

ATM c.3154_splice (p.A1052_splice), exon 22 - in 3% of 274 reads***

CHEK2 c.335A>G (p.N112S), exon 3 - in 54% of 247 reads***

DMD c.6058G>C (p.A2020P), exon 42 - in 98% of 174 reads***

GATA3 c.1294C>A (p.P432T), exon 6 - in 4% of 294 reads***

GLI2 c.1585G>A (p.G529R), exon 10 - in 51% of 19 4 reads***

KDR c.1082A>G (p.E361G), exon 8 - in 55% of 276 reads***

NTRK1 c.1888G>T (p.V630L), exon 15 - in 4% of 119 reads***

PDGFRA c.1045T>A (p.W349R), exon 7 - in 3% of 359 reads***

SOX9 c.920C>A (p.P307Q), exon 3 - in 3% of 232 reads***

SUZ12 c.698C>G (p.S233C), exon 7 - in 42% of 238 reads***

COPY NUMBER VARIATIONS:

1p36.13	SDHB	Single copy deletion
9q34.1	ABL1	Low copy number gain
9q34.3	NOTCH1	Low copy number gain

INTERPRETATION:

One or more mutations are detected in this sample at an allele fraction of 50% or 100%, which is suspicious for a germline alteration that has no significance for cancer biology or treatment, especially given the estimated tumor percentage of 10%. Moreover, these alterations have not been previously reported in a cancer sample. However, these variations have not been reported in databases of known germline alterations and, therefore, it is not possible to definitively characterize them as benign. Nevertheless, they should be interpreted with caution.

Several mutations are reported in the 3-5% range, which is below our threshold for reporting (10%), but are included here because the tumor content was so low.

Tumor content was too low for accurate assessment of copy number, particularly with regard to low level amplifications or single copy losses. Please interpret with caution.

Fundamentally, this sample did not contain enough tumor cells to be suitable for this analysis.

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TEST INFORMATION:

BACKGROUND:

Somatic genetic alterations in oncogenes and tumor-suppressor genes contribute to the pathogenesis and evolution of human cancers. These alterations can provide prognostic and predictive information and stratify cancers for targeted therapeutic information. We classify these alterations into five tiers using the following guidelines:

Tier 1: The alteration has well-established published evidence confirming clinical utility in this tumor type, in at least one of the following contexts: predicting response to treatment with an FDA-approved therapy; assessing prognosis; establishing a definitive diagnosis; or conferring an inherited increased risk of cancer to this patient and family.

Tier 2: The alteration may have clinical utility in at least one of the following contexts: selection of an investigational therapy in clinical trials for this cancer type; limited evidence of prognostic association; supportive of a specific diagnosis; proven association of response to treatment with an FDA-approved therapy in a different type of cancer; or similar to a different mutation with a proven association with response to treatment with an FDA-approved therapy in this type of cancer.

Tier 3: The alteration is of uncertain clinical utility, but may have a role as suggested by at least one of the following: demonstration of association with response to treatment in this cancer type in preclinical studies (e.g., in vitro studies or animal models); alteration in a biochemical pathway that has other known, therapeutically-targetable alterations; alteration in a highly conserved region of the protein predicted, in silico, to alter protein function; or selection of an investigational therapy for a different cancer type.

Tier 4: The alteration is novel or its significance has not been studied in cancer.

Tier 5: The alteration has been determined to have no clinical utility, either for selecting therapy, assessing prognosis, establishing a diagnosis, or determining hereditary disease risk.

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METHODOLOGY:

We have developed a cancer genomic assay to detect somatic mutations, copy number variations and structural variants in tumor DNA extracted from fresh, frozen or formalin-fixed paraffin-embedded samples. The OncoPanel assay surveys exonic DNA sequences of 299 cancer genes and 113 introns across 35 genes for rearrangement detection. DNA is isolated from tissue containing at least 20% tumor nuclei and analyzed by massively parallel sequencing using a solution-phase Agilent SureSelect hybrid capture kit and an Illumina HiSeq 2500 sequencer.

The 299 genes are: ABL1, AKT1, AKT2, AKT3, ALK, ALOX12B, APC, AR, ARAF, ARID1A, ARID1B, ARID2, ASXL1, ATM, ATRX, AURKA, AURKB, AXL, B2M, BAP1, BCL2, BCL2L1, BCL2L12, BCL6, BCOR, BCORL1, BLM, BMPR1A, BRAF, BRCA1, BRCA2, BRD4, BRIP1, BUB1B, CADM2, CARD11, CBL, CBLB, CCND1, CCND2, CCND3, CCNE1, CD274, CD58, CD79B, CDC73, CDH1, CDK1, CDK2, CDK4, CDK5, CDK6, CDK9, CDKN1A, CDKN1B, CDKN1C, CDKN2A, CDKN2B, CDKN2C, CEBPA, CHEK2, CIITA, CREBBP, CRKL, CRLF2, CRTC1, CRTC2, CSF1R, CSF3R, CTNNB1, CUX1, CYLD, DDB2, DDR2, DEPDC5, DICER1, DIS3, DMD, DNMT3A, EED, EGFR, EP300, EPHA3, EPHA5, EPHA7, ERBB2, ERBB3, ERBB4, ERCC2, ERCC3, ERCC4, ERCC5, ESR1, ETV1, ETV4, ETV5, ETV6, EWSR1, EXT1, EXT2, EZH2, FAM46C, FANCA, FANCC, FANCD2, FANCE, FANCF, FANCG, FAS, FBXW7, FGFR1, FGFR2, FGFR3, FGFR4, FH, FKBP9, FLCN, FLT1, FLT3, FLT4, GATA3, GATA4, GATA6, GLI1, GLI2, GLI3, GNA11, GNAQ, GNAS, GNB2L1, GPC3, GSTM5, H3F3A, HNF1A, HRAS, ID3, IDH1, IDH2, IGF1R, IKZF1, IKZF3, INSIG1, JAK2, JAK3, KCNIP1, KDM5C, KDM6A, KDM6B, KDR, KEAP1, KIT, KRAS, LINC00894, LMO1, LMO2, LMO3, MAP2K1, MAP2K4, MAP3K1, MAPK1, MCL1, MDM2, MDM4, MECOM, MEF2B, MEN1, MET, MITF, MLH1, MLL, MLL2, MPL, MSH2, MSH6, MTOR, MUTYH, MYB, MYBL1, MYC, MYCL1, MYCN, MYD88, NBN, NEGR1, NF1, NF2, NFE2L2, NFKBIA, NFKBIZ, NKX2-1, NOTCH1, NOTCH2, NPM1, NPRL2, NPRL3, NRAS, NTRK1, NTRK2, NTRK3, PALB2, PARK2, PAX5, PBRM1, PDCD1LG2, PDGFRA, PDGFRB, PHF6, PHOX2B, PIK3C2B, PIK3CA, PIK3R1, PIM1, PMS1, PMS2, PNRC1, PRAME, PRDM1, PRF1, PRKAR1A, PRKCI, PRKCZ, PRKDC, PRPF40B, PRPF8, PSMD13, PTCH1, PTEN, PTK2, PTPN11, PTPRD, QKI, RAD21, RAF1, RARA, RB1, RBL2, RECQL4, REL, RET, RFWD2, RHEB, RHPN2, ROS1, RPL26, RUNX1, SBDS, SDHA, SDHAF2, SDHB, SDHC, SDHD, SETBP1, SETD2, SF1, SF3B1, SH2B3,

SLITRK6, SMAD2, SMAD4, SMARCA4, SMARCB1, SMC1A, SMC3, SMO, SOCS1, SOX2, SOX9, SQSTM1, SRC, SRSF2, STAG1, STAG2, STAT3, STAT6, STK11, SUFU, SUZ12, SYK, TCF3, TCF7L1, TCF7L2, TERC, TERT, TET2, TLR4, TNFAIP3, TP53, TSC1, TSC2, U2AF1, VHL, WRN, WT1, XPA, XPC, XPO1, ZNF217, ZNF708, ZRSR2.

Intronic regions are tiled on specific introns of ABL1, AKT3, ALK, BCL2, BCL6, BRAF, CIITA, EGFR, ERG, ETV1, EWSR1, FGFR1, FGFR2, FGFR3, FUS, IGH@, IGK@, IGL@, JAK2, MLL, MYC, NPM1, NTRK1, PAX5, PDGFRA, PDGFRB, PPARG, RAF1, RARA, RET, ROS1, SS18, TRA@, TRB@, TRG@.TMPRSS2.

For detailed methodology and protocol, please contact the Center for Advanced Molecular Diagnostics (857-307-1500).

These tests were developed and their performance characteristics determined by the Molecular Diagnostics Laboratory, Brigham and Women's Hospital. They have 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.

References:

Wagle et al. High-throughput detection of actionable genomic alterations in clinical tumor samples by targeted, massively parallel sequencing. Cancer Discov. 2012 Jan;2(1):82-93.

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