OncoStrands™ Comprehensive Panel

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PATIENT		PHYSICIAN	SPECIMEN	CASE
NAME P F	**SEX** Male	**ORDERING PHYSICIAN** Dr XXXXX XXXXXXXXXXX	**EXT. SPECIMEN ID** XXXXXXX	**ACCESSION#** A23MXXXX
DATE OF BIRTH 25/11/1900	**MRN#** -	**FACILITY** XXXX Medical	**DATE RECEIVED** 09/02/2023	**DATE REPORTED** 17/02/2023
DISEASE Cholangiocarcinoma of biliary tract		**DATE ORDERED** 01/02/2023	**SPECIMEN TYPE** Formalin-fixed paraffin-embedded tissue specimen	**REVIEW STATUS** Final
ADDRESS -		**COPY TO** -	**% TUMOR CELLULARITY** 80%	**REPORTED BY** Dr. Vivek Rathi

Report Summary

• A fusion transcript for FGFR2 gene detected –

TIER 1A , ARF3-FGFR2 fusion – currently, FGFR inhibitors like pamigatinib, infrigatinib and futibatinib are FDA approved for cholangiocarcinoma patients harbouring FGFR2 fusions.

• Tumour mutation burden (TMB) status – HIGH. Patients with any solid tumour demonstrating a high (>10 Muts/Mb) TMB are known to benefit from immune checkpoint inhibitor pembrolizumab.

• Microsatellite instability (MSI) status – Stable.

• Pertinent negatives –
Mutations – no pathogenic variants detected in IDH1, BRAF, BRCA1, BRCA2, HER2, or FGFR2.

Fusions – no fusion transcripts detected in NTRK1, NTRK2 or NTRK3.

IA	IB	IIC	IID	TMB	MSI	HRD	Trials
1	0	1	0	High 14.1 Mut/Mb	Stable (1.6% Unstable Sites)	Not Tested N/A Genomic Instability Score (GIS)	0

Clinical Implications

TIER	VARIANT DETECTED (GENE/SYNTAX)	CLINICAL IMPACT		SELECT CLINICAL TRIALS
IA	**ARF3, FGFR2** ARF3, FGFR2 fusion transcript	**May benefit from:**	Pamigatinib, Infrigatinib, Futibatinib	0
		In Tumor Type:	Cholangiocarcinoma	

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TIER	VARIANT DETECTED (GENE/SYNTAX)	CLINICAL IMPACT	SELECT CLINICAL TRIALS
IIC	TP53 p.S215G	No guidelines existing in the report.	0

Other Biomarkers

BIOMARKER	RESULT	CLINICAL IMPACT
TMB	High 14.1 muts/Mb	Tumour mutation burden (TMB) or mutational load, is a measure of the number of somatic protein-coding base substitution and insertion/ deletion mutations in a tumour specimen. TMB varies between tumour types and can range between very low to very high, and is affected by a variety of causes including microsatellite instability (MSI, PMID: 22810696, 23636398), mutations in the proof reading domains of DNA polymerases encoded by the POLE and POLD1 genes (PMID: 25568919), or exposure to environmental mutagens like exposure to ultraviolet light in melanoma (PMID: 23875803), or cigarette smoking in lung cancer (PMID: 25765070). Multiple clinical trials of immune checkpoint (PD-1/ PD-L1) inhibitors in different tumour types have reported that patients with TMB of less than 10 Muts/Mb do not derive as much clinical benefits from these agents than those with TMB equal to or greater than 10 Muts/ Mb. In the phase II basket study KEYNOTE-158 (NCT02628067), of 102 patients with TMB-H tumours given pembrolizumab, 30 responded (ORR 29%, 95% CI, 21-39) with 4% having a complete response and 50% having ongoing responses after 24 months. In June 2020, US FDA gave accelerated approval for pembrolizumab in patients with unresectable or metastatic solid tumours that have progressed on prior treatment or have no other treatment options. This sample harbours a TMB levels known to be associated with sensitivity to PD-1 or PDL-1 targeting immune checkpoint inhibitors and may benefit from Pembrolizumab.
MSI	Stable 1.6% Unstable Sites	Microsatellite instability (MSI) is a condition that generates excessive amount of short insertions or deletion mutations in the genome and is caused by a deficiency in DNA mismatch repair (MMR) in the tumour (PMID: 26337942). This condition occurs due to genetic or epigenetic inactivation of one of the MMR pathway proteins, primarily MLH1, PMS2, MSH2, or MSH6 (PMID: 21081928). Microsatellite Stable (MSS) status indicates a MMR proficient cancer, and generally correlates with intact expression of all MMR family proteins (PMID:15528785). Clinical evidence suggests that MSS tumours compared to MicroSatellite Instability High

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BIOMARKER	RESULT	CLINICAL IMPACT
		(MSI-H) tumours, are less likely to respond to immune checkpoint inhibitor therapies with pembrolizumab or nivolumab.
HRD	**Not Tested** N/A Genomic Instability Score (GIS)	Genomic instability status not tested for this sample.

Other Test Results

- Histopathology -
  Liver biopsy - metastatic moderately differentiated adenocarcinoma.

Clinical Interpretations

ARF3, FGFR2	**ARF3, FGFR2 fusion transcript**		Tier IA		VAF:	Depth:

GENE: FGFR2, fibroblast growth factor receptor 2, is a receptor tyrosine kinase activated upon binding of the FGF ligand, which activates RAS-MAPK and PI3K-AKT pathways (PMID: 22508544). Altered function of FGFR2 through activating mutations, fusions, and amplification increases cell proliferation and tumorigenesis (PMID: 22508544) and is observed in prostate (PMID: 30761180), breast, lung, uterine, and ovarian cancers (PMID: 29104507), while FGFR2 amplification (PMID: 31076567) and overexpression (PMID: 30662521) is commonly observed in gastric cancer.

VARIANT: A fusion between genes ARF3 and FGFR2 detected; breakpoint - t(10;12)(q26;q13)(chr10:g.123239535::chr12:g.49333438).

THERAPEUTICS: Currently, FGFR inhibitors like pamigatinib, infrigatinib and futibatinib are FDA approved for treatment of patients with cholangiocarcinoma harbouring FGFR2 fusion.

In a Phase II trial that supported FDA approval, Truseltiq (infigratinib) treatment demonstrated manageable toxicity, resulted in an objective response rate of 23.1% (25/108, 1 complete response, 24 partial responses) in patients with previously treated advanced cholangiocarcinoma harboring an FGFR2 fusion or rearrangement, with a median duration of response of 5.0 months and a median progression-free survival of 7.3 months (J Clin Oncol 39, no. 3_suppl (January 20, 2021) 265-265; NCT02150967).In a Phase II (FIGHT-202) trial, Pemazyre (pemigatinib) treatment resulted in an objective response in 35.5% (38/107, 3 complete response, 35 partial response) of patients with advanced cholangiocarcinoma harboring FGFR2 fusions or rearrangements, with a disease control rate of 82% (88/107), median time-to-response of 2.7 months, and a median progression-free survival of 6.9 months (PMID: 32203698; NCT02924376).In a Phase II trial (FOENIX-CCA2) that supported FDA approval, Lytgobi (futibatinib) demonstrated a manageable toxicity profile and resulted in an objective response rate of 41.7% (43/103), a disease control rate of 82.5% in patients with advanced or metastatic intrahepatic cholangiocarcinoma harboring FGFR2 fusions or other rearrangements, with a median progression-free survival of 8.9 mo and median overall survival of 20.0 mo (J Clin Oncol 40, no. 16_suppl (June 01, 2022) 4009; NCT02052778).

TP53	**p.S215G**	c.643A>G	Tier IIC	NM_001126114.2	VAF: 72.5%	Depth: 829

GENE: TP53, tumor protein p53, is a tumor suppressor (PMID: 30562755) and oncogene (PMID: 30577483) involved in cell cycle arrest and apoptosis, and is the most frequently mutated gene in cancer (PMID: 10065147, PMID: 22713868). TP53 germline mutations are common in Li-Fraumeni syndrome (PMID: 30239254) and somatic missense mutations are frequent in almost all cancer types (PMID: 30224644) and are also implicated in chemoresistance (PMID: 9927204, PMID: 24065105, PMID: 27066457).

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VARIANT: A missense mutation, p.S215G (c.643A>G), was identified in the TP53 gene. This specific mutation resides in the P53 DNA-binding domain (95-288) and results in decreased TP53 activity and reduced apoptosis relative to wild type TP53 in cell culture (PMID: 22862161). TP53, tumor protein p53, is a tumor suppressor (PMID: 30562755) and oncogene (PMID: 30577483) involved in cell cycle arrest and apoptosis, and is the most frequently mutated gene in cancer (PMID: 10065147, PMID: 22713868).

THERAPEUTICS: Currently, there are no FDA approved or NCCN-Compendium recommended treatment options for cholangiocarcinoma patients harbouring this variant.

In a Phase I trial, the combination of Votrient (pazopanib) and Zolinza (vorinostat) improved progression-free survival and overall survival in advanced solid tumor patients harboring TP53 hotspot mutations, and resulted in an increased stable disease rate of 45% (5/11), compared to a stable disease rate of 16% (4/25) in patients without detected TP53 mutations (PMID: 25669829). In a Phase I trial, Adavosertib (MK-1775) treatment resulted in a prtial response in 3 and progressive disease in 2 of 6 patients with advanced solid tumors harboring TP53 mutations (J Clin Oncol 38: 2020 (suppl; abstr 3624); NCT01748825).In a retrospective analysis of a Phase I trial, Adavosertib (MK-1775) combined with a chemotherapy resulted in a 21% (4/19) response rate in advanced solid tumor patients harboring a TP53 mutation and in those without a TP53 mutation, a 12% (4/33) response rate was observed (PMID: 27601554). In a clinical study, VEGF/VEGFR inhibitor treatment resulted in improved rates of response (stable disease over 6 months/partial/complete response, 31% vs 7%), time-to-treatment failure, and overall survival (both $p<0.01$) compared to control in patients with TP53 mutant advanced solid tumors (n=106), but not in patients with TP53 wild-type tumors (n=82) (PMID: 27466356). In a retrospective study, Avastin (bevacizumab) treatment was associated with increased progression-free survival in cancer patients carrying TP53 mutations (PMID: 23670029).

Clinical Trials

No relevant clinical trials were reported.

Significant Negative Findings

As reported in the Summary section above.

Tier III - Variants of Uncertain Significance

AR	*BARD1*	*BARD1*	*CCND3*	*CCNE1*
p.Q196L NM_000044.3 c.587A>T VAF 37.5 % DEPTH 858	p.? NM_000465.2 c.1678-1G>A VAF 42.8 % DEPTH 965	p.P418H NM_000465.2 c.1253C>A VAF 38.3 % DEPTH 1,607	p.S259A NM_001760.3 c.774_775delinsTG VAF 44.6 % DEPTH 489	Copy number gain in *CCNE1* (50 copies)

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CD276
p.T160M
NM_001024736.1
c.471_479delins9
VAF 49.9 %
DEPTH 830

CIC
p.G1012R
NM_015125.3
c.3034G>A
VAF 54.5 %
DEPTH 697

DDR2
p.K59E
NM_001014796.1
c.175A>G
VAF 30.1 %
DEPTH 1,401

DOT1L
p.V1236I
NM_032482.2
c.3706G>A
VAF 13.6 %
DEPTH 647

EPHA5
p.V426I
NM_004439.5
c.1276G>A
VAF 48.5 %
DEPTH 1,231

ERCC2
p.V714Gfs
NM_000400.3
c.2133_2144delinsTGAG
VAF 28.4 %
DEPTH 675

FAT1
p.E1602G
NM_005245.3
c.4805A>G
VAF 34.5 %
DEPTH 1,124

FAT1
p.A173T
NM_005245.3
c.517G>A
VAF 8.4 %
DEPTH 1,599

FGFR4
p.G388R
NM_002011.3
c.1162G>A
VAF 45.4 %
DEPTH 610

GNAS
p.(=)
NM_080425.2
c.1677C>T
VAF 49.5 %
DEPTH 1,247

HFM1, ETV1
HFM1-ETV1 fusion transcript

HSD3B1
p.W4*
NM_000862.2
c.12G>A
VAF 33.6 %
DEPTH 746

KDR
p.Q472H
NM_002253.2
c.1416A>T
VAF 48.8 %
DEPTH 767

LATS1
p.N271D
NM_004690.3
c.811A>G
VAF 35.2 %
DEPTH 1,115

LRP1B
p.T4519A
NM_018557.2
c.13555A>G
VAF 84.3 %
DEPTH 654

MCL1
p.A227V
NM_021960.4
c.680C>T
VAF 23 %
DEPTH 2,324

NBN
p.E248K
NM_002485.4
c.742G>A
VAF 41.9 %
DEPTH 2,368

PREX2
p.S1530N
NM_024870.2
c.4589G>A
VAF 51.3 %
DEPTH 1,815

PRKDC
p.V2937I
NM_006904.6
c.8809G>A
VAF 14 %
DEPTH 1,938

PTCH1
p.L4F
VAF 5.5 %
DEPTH 578

QKI
p.A239T
NM_006775.2
c.715G>A
VAF 38.7 %
DEPTH 905

ROS1
p.K2228_S2229delinsQC
NM_002944.2
c.6682_6686delins5
VAF 45.8 %
DEPTH 718

ROS1
p.K831T
NM_002944.2
c.2492A>C
VAF 33.4 %
DEPTH 876

SOX17
p.P226L
NM_022454.3
c.677C>T
VAF 16.2 %
DEPTH 853

SPEN
p.P2067L
NM_015001.2
c.6200C>T
VAF 63.5 %
DEPTH 1,138

VHL
p.L184F
NM_000551.3
c.550C>T
VAF 34.4 %
DEPTH 791

Other Comments

FDA-approved treatments for specific genetic alterations in cholangiocarcinoma -

IDH1 mutations - Ivosidenib.
FGFR2 fusions - Pemigatinib, infigratinib.

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FDA-approved treatments for specific biomarkers in tumor type–agnostic indications –

NTRK1 or NTRK2 or NTRK3 fusions - Entrectinib, larotrectinib.
MSI-H, TMB-H - Pembrolizumab.

FDA-listed genetic alterations contraindicated for specific treatments –

NTRK1 and NTRK3 known acquired resistance mutations (eg, NTRK1 G595R and G667C; NTRK3 F617L, G623R, and G696A) - Entrectinib, larotrectinib.

NCCN Biomarkers Compendium recommended genes for testing in cholangiocarcinoma –

Mutations - BRAF, BRCA1, BRCA2, ERBB2, FGFR2, IDH1
Fusions - NTRK1, NTRK2, NTRK3, RET
Other biomarkers - MSI/MMR, TMB

Tier Definitions

Tier I-A: Approved therapy. Included in professional guidelines.
Tier I-B: Well-powered studies with consensus from experts in the field.
Tier II-C: Approved therapies for different tumour types or investigational therapies. Multiple small published studies with some consensus. Inclusion criteria for clinical trials.
Tier II-D: Limited clinical or preclinical studies.
Tier III (VUS): Variants of Unknown Clinical Significance.
Tier IV: Benign or likely benign variants (not included in the report)

Test Information

REPORTED GENES: A total of 523 genes were subjected to targeted next generation sequencing analysis. Details available upon request. **CGW VERSION:** CGW_v6.21.0.1 **DATABASE DETAILS:** The versions, releases, builds, dates of the following databases were used to generate this report: Genomic Build: GRCh37.p13 | Genomic Annotation Sources: NCBI RefSeq v105 | gnomAD: r2.1 | dbNSFP: 4.3c | COSMIC: v96 | ExAC: v1.0 | NHLBI ESP: v.0.0.30 | dbSNP: 149 | ClinVar: 20221001 **ASSAY METHODOLOGY:**

PATHOLOGY ASSESSMENT: Pathologist reviews on H&E stained section of the tissue block or stained cytology slide were considered to assess adequacy and if needed, guide enrichment of tumour for sequencing analysis. The in-house validation ensured that the samples passed all established laboratory QC metrics.

ASSAY METHODS: This is an in-house IVD test and has been extensively validated using the Illumina TruSight™ Oncology 500 (TSO500) targeted hybridcapture based next generation sequencing chemistry. It employs Unique molecular identifiers (UMI) to enable detection of variants, present in formalin-fixed paraffin-embedded (FFPE) tumor samples, at low VAFs with a high degree of sensitivity and specificity. TSO500 is designed to detect multiple classes of mutations including single-nucleotide variants (SNVs), multi-nucleotide variants (<3bp), small Insertions (1-18 bp) Deletions (1-27 bp) and Copy Number Variants (CNVs). The assay also detects, quantitatively, microsatellite instability (MSI) and tumour mutational burden (TMB). Genomic Instability Score (HRD) is an optional component of this test and is only performed upon request and the result is added in the 'Other Biomarkers' section of the report. Fusions and splice variants are detected in RNA. DNA and RNA were extracted from the same FFPE tissue, and the RNA was then reverse transcribed to cDNA. The genomic DNA was ultrasonically sheared to prepare sequencing libraries. The genomic regions of interest were hybridized to biotinylated probes, magnetically pulled down with streptavidin-coated beads, and eluted to enrich the library pool. Finally, NGS libraries were normalized using a simple bead-based protocol, then pooled and sequenced on an Illumina NextSeq™ 550 instrument.

SECONDARY ANALYSIS METHODS: The DNA and RNA data is analysed using the Illumina Software TSO500 v2.2 Local App and a customized analysis pipeline within the Clinical Genomics Workspace software platform from PierianDx.

VARIANT CALLING: Variants were reported according to the HGVS nomenclature (www.hgvs.org/mutnomen) and classified as per the AMP classification system into tiers IA, IB, IIC, IID, III and IV. These tiers are stratified by clinical utility ('actionability' for clinical decision-making as to diagnosis, prognosis, treatment options, and carrier status) and previously reported data in the medical literature. Variations found in

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gnomAD (https:gnomad.broadinstitute.org) that have ≥1% minor allele frequency (except those that are also in ClinVar denoted as clinically relevant, used in a clinical diagnostic assay, or reported as a mutation in a publication) are classified as known polymorphisms. Exons from some transcripts included in the RefSeq annotation release v105 found in genes reported in certain gene subsets of this test are not targeted by the assay. Some variants in genes with sequence homology to multiple genomic locations are not reported. This assay does not detect complex indels and complex structural alterations. Variants located outside of the targeted regions will not be detected. It is possible that pathogenic variants may not be reported by one or more of the tools because of the parameters used. However, tool parameters were optimized to maximize specificity and sensitivity. The assay cannot differentiate between somatic and germline variants. CNV gain was analysed using the following criteria: Not Detected (≤5 copies), Equivocal (>5, <10), and Detected (≥10). The assay is not validated to detect copy number loss. The thresholds of ≥20%MSI unstable sites and ≥10 Mut/Mb were used to call MSI unstable and TMB high tumors, respectively. Genomic Instability Score (GIS) is a whole genome signature for homologous recombination deficiency. The GIS is composed of the sum of three components: loss of heterozygosity, telomeric allele imbalance, and large-scale state transition. These components are estimated using the GIS algorithm obtained by the Manufacturer from Myriad Genetics, which uses an input of the b-allele frequency and coverage across a genome-wide single nucleotide panel. A panel of normal samples is used for both bias reduction and normalization prior to GIS estimation.

OncoStrands™Comprehensive Panel Gene List

Small variants (DNA): *ABL1 ABL2 ACVR1 ACVR1B AKT1 AKT2 AKT3 ALK ALOX12B ANKRD11 ANKRD26 APC AR ARAF ARFRP1 ARID1A ARID1B ARID2 ARID5B ASXL1 ASXL2 ATM ATR ATRX AURKA AURKB AXIN1 AXIN2 AXL B2M BAP1 BARD1 BBC3 BCL10 BCL2 BCL2L1 BCL2L11 BCL2L2 BCL6 BCOR BCORL1 BCR BIRC3 BLM BMPR1A BRAF BRCA1 BRCA2 BRD4 BRIP1 BTG1 BTK C11orf30 CALR CARD11 CASP8 CBFB CBL CCND1 CCND2 CCND3 CCNE1 CD274 CD276 CD74 CD79A CD79B CDC73 CDH1 CDK12 CDK4 CDK6 CDK8 CDKN1A CDKN1B CDKN2A CDKN2B CDKN2C CEBPA CENPA CHD2 CHD4 CHEK1 CHEK2 CIC CREBBP CRKL CRLF2 CSF1R CSF3R CSNK1A1 CTCF CTLA4 CTNNA1 CTNNB1 CUL3 CUX1 CXCR4 CYLD DAXX DCUN1D1 DDR2 DDX41 DHX15 DICER1 DIS3 DNAJB1 DNMT1 DNMT3A DNMT3B DOT1L E2F3 EED EGFL7 EGFR EIF1AX EIF4A2 EIF4E EML4 EP300 EPCAM EPHA3 EPHA5 EPHA7 EPHB1 ERBB2 ERBB3 ERBB4 ERCC1 ERCC2 ERCC3 ERCC4 ERCC5 ERG ERRFI1 ESR1 ETS1 ETV1 ETV4 ETV5 ETV6 EWSR1 EZH2 FAM123B FAM175A FAM46C FANCA FANCC FANCD2 FANCE FANCF FANCG FANCI FANCL FAS FAT1 FBXW7 FGF1 FGF10 FGF14 FGF19 FGF2 FGF23 FGF3 FGF4 FGF5 FGF6 FGF7 FGF8 FGF9 FGFR1 FGFR2 FGFR3 FGFR4 FH FLCN FLI1 FLT1 FLT3 FLT4 FOXA1 FOXL2 FOXO1 FOXP1 FRS2 FUBP1 FYN GABRA6 GATA1 GATA2 GATA3 GATA4 GATA6 GEN1 GID4 GLI1 GNA11 GNA13 GNAQ GNAS GPR124 GPS2 GREM1 GRIN2A GRM3 GSK3B H3F3A H3F3B H3F3C HGF HIST1H1C HIST1H2BD HIST1H3A HIST1H3B HIST1H3C HIST1H3D HIST1H3E HIST1H3F HIST1H3G HIST1H3H HIST1H3I HIST1H3J HIST2H3A HIST2H3C HIST2H3D HIST3H3 HLA-A HLA-B HLA-C HNF1A HNRNPK HOXB13 HRAS HSD3B1 HSP90AA1 ICOSLG ID3 IDH1 IDH2 IFNGR1 IGF1 IGF1R IGF2 IKBKE IKZF1 IL10 IL7R INHA INHBA INPP4A INPP4B INSR IRF2 IRF4 IRS1 IRS2 JAK1 JAK2 JAK3 JUN KAT6A KDM5A KDM5C KDM6A KDR KEAP1 KEL KIF5B KIT KLF4 KLHL6 KMT2B KMT2C KMT2D KRAS LAMP1 LATS1 LATS2 LMO1 LRP1B LYN LZTR1 MAGI2 MALT1 MAP2K1 MAP2K2 MAP2K4 MAP3K1 MAP3K13 MAP3K14 MAP3K4 MAPK1 MAPK3 MAX MCL1 MDC1 MDM2 MDM4 MED12 MEF2B MEN1 MET MGA MITF MLH1 MLL MLLT3 MPL MRE11A MSH2 MSH3 MSH6 MST1 MST1R MTOR MUTYH MYB MYC MYCL1 MYCN MYD88 MYOD1 NAB2 NBN NCOA3 NCOR1 NEGR1 NF1 NF2 NFE2L2 NFKBIA NKX2-1 NKX3-1 NOTCH1 NOTCH2 NOTCH3 NOTCH4 NPM1 NRAS NRG1 NSD1 NTRK1 NTRK2 NTRK3 NUP93 NUTM1 PAK1 PAK3 PAK7 PALB2 PARK2 PARP1 PAX3 PAX5 PAX7 PAX8 PBRM1 PDCD1 PDCD1LG2 PDGFRA PDGFRB PDK1 PDPK1 PGR PHF6 PHOX2B PIK3C2B PIK3C2G PIK3C3 PIK3CA PIK3CB PIK3CD PIK3CG PIK3R1 PIK3R2 PIK3R3 PIM1 PLCG2 PLK2 PMAIP1 PMS1 PMS2 PNRC1 POLD1 POLE PPARG PPM1D PPP2R1A PPP2R2A PPP6C PRDM1 PREX2 PRKAR1A PRKCI PRKDC PRSS8 PTCH1 PTEN PTPN11 PTPRD PTPRS PTPRT QKI RAB35 RAC1 RAD21 RAD50 RAD51 RAD51B RAD51C RAD51D RAD52 RAD54L RAF1 RANBP2 RARA RASA1 RB1 RBM10 RECQL4 REL RET RFWD2 RHEB RHOA RICTOR RIT1 RNF43 ROS1 RPS6KA4 RPS6KB1 RPS6KB2 RPTOR RUNX1 RUNX1T1 RYBP SDHA SDHAF2 SDHB SDHC SDHD SETBP1 SETD2 SF3B1 SH2B3 SH2D1A SHQ1 SLIT2 SLX4 SMAD2 SMAD3 SMAD4 SMARCA4 SMARCB1 SMARCD1 SMC1A SMC3 SMO SNCAIP SOCS1 SOX10 SOX17 SOX2 SOX9 SPEN SPOP SPTA1 SRC SRSF2 STAG1 STAG2 STAT3 STAT4 STAT5A STAT5B STK11 STK40 SUFU SUZ12 SYK TAF1 TBX3 TCEB1 TCF3 TCF7L2 TERC TERT TET1 TET2 TFE3 TFRC TGFBR1 TGFBR2 TMEM127 TMPRSS2 TNFAIP3 TNFRSF14 TOP1 TOP2A TP53 TP63 TRAF2 TRAF7 TSC1 TSC2 TSHR U2AF1 VEGFA VHL VTCN1 WISP3 WT1 XIAP XPO1 XRCC2 YAP1 YES1 ZBTB2 ZBTB7A ZFHX3 ZNF217 ZNF703 ZRSR2*

Amplifications (DNA): *AKT2 ALK AR ATM BRAF BRCA1 BRCA2 CCND1 CCND3 CCNE1 CDK4 CDK6 CHEK1 CHEK2 EGFR ERBB2 ERBB3 ERCC1 ERCC2 ESR1 FGF1 FGF10 FGF14 FGF19 FGF2 FGF23 FGF3 FGF4 FGF5 FGF6 FGF7 FGF8 FGF9 FGFR1 FGFR2 FGFR3 FGFR4 JAK2 KIT KRAS LAMP1 MDM2 MDM4 MET MYC MYCL1 MYCN NRAS NRG1 PDGFRA PDGFRB PIK3CA PIK3CB PTEN RAF1 RET RICTOR RPS6KB1 TFRC*

Fusions (RNA): *ABL1 AKT3 ALK AR AXL BCL2 BRAF BRCA1 BRCA2 CDK4 CSF1R EGFR EML4 ERBB2 ERG ESR1 ETS1 ETV1 ETV4 ETV5 EWSR1 FGFR1 FGFR2 FGFR3 FGFR4 FLI1 FLT1 FLT3 JAK2 KDR KIF5B KIT MET MLL MLLT3 MSH2 MYC NOTCH1 NOTCH2 NOTCH3 NRG1 NTRK1 NTRK2 NTRK3 PAX3 PAX7 PDGFRA PDGFRB PIK3CA PPARG RAF1 RET ROS1 RPS6KB1 TMPRSS2*

DISCLAIMER:

This is a laboratory developed test, and its performance characteristics have been determined by LifeStrands Genomics. Variants below the limit of detection, insertions/deletions >40 bp, and fusions that do not alter expressed messenger RNA may not be detected by this assay. A negative result does not exclude the presence of a variant beyond these detection limitations. The assay is not informative for mutations outside the 523 cancer-related genes or for those regions for which the assay achieves limited coverage. This assay is not validated for large (>40 bp) indels, complex structural variants, or gene-level deletion events. Further, the assay is not validated to detect fusions that do not alter the expressed transcript (such as those that occur in the MYC gene). MSI analysis can identify MSI Positive and MSI Negative; however, MSI-Low may not be detected.

PATIENT	DOB	DISEASE	ACCESSION	MRN	REPORT DATE	REPORT STATUS
P F	25/11/1900	Cholangiocarcinoma of biliary tract	A23MXXXX	-	17/02/2023	Final

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Report electronically reviewed and signed out by: Dr. Vivek Rathi
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