An End-To-End Precision Medicine Approach for Matching Cancer Patients to Clinical Trials

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INTRODUCTION

Precise matching of a patient's tumor genomic profile with appropriate targeted agents is the goal of precision medicine, and the salutary benefits of these targeted therapies upon survival has been well documented in the literature. Using a 413+ cancer-related actionable gene, NGS sequencing panel, KEW CancerPlex®, we analyzed a variety of solid tumors to identify mutations associated with predicted response to approved and investigational targeted therapy agents.

METHODS

Tumor profiling was conducted in a CAP/CLIA-certified laboratory, licensed by MA and 48 other states (KEW Group Inc.). DNA was extracted from FFPE tissue sections, slides, cell blocks, from FNAs or effusions, or cell pellets, followed by hybrid-capture next-gen sequencing. Rapid sequencing runs were employed to generate at least 200x depth, and mutational analysis was performed using the KEW Clinical Genomics Analytical pipeline. The coding regions and portions of the introns of 413+ genes were sequenced. The assay simultaneously surveyed multiple classes of genomic abnormalities including single nucleotide substitutions (SNP), small insertions/deletions (indels), copy number alterations (CNV), and translocations.

Variant characterization and annotation were conducted for a MAF cut-off of 10%. Genotype-based personalized molecular modeling was performed to characterize tumors, and response or resistance to FDA-approved drugs was predicted. Matches were identified by screening 260,000 clinical trials, including approximately sixty thousands active or recruiting studies.

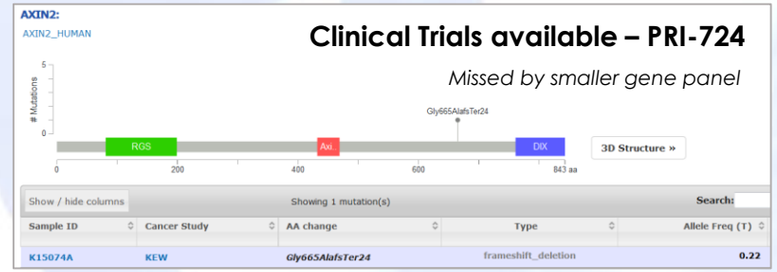
TECHNICAL PERFORMANCE - CancerPlex® v3

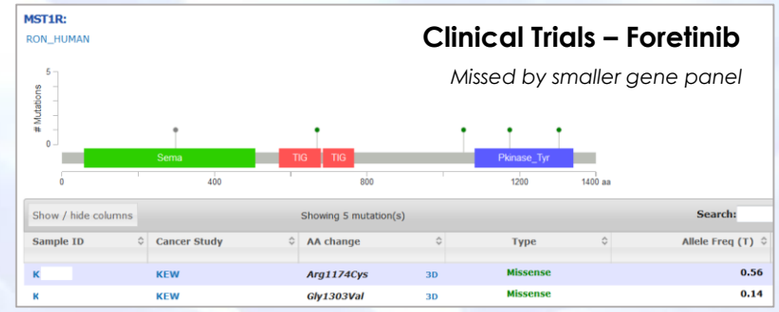
Analytic sensitivity for SNP calls is 98.9%-100% at 95% confidence interval, specificity is 99.99%; for indels sensitivity is 99.8%-100% at 95% confidence interval and specificity is 99.99%. Analytic sensitivity for translocations of the *ALK*, *RET*, and *ROS1* genes is 100% for tumor purity as low as 10%.
Testing and reporting turnaround time of 7-10 days.
CancerPlex Test requires only a small portion of the biopsy (40 micron section or equivalency), and it can technically be performed with a very low amount of input DNA. The CancerPlex requirement of tissue is determined by biological considerations to obtain a representative sampling of the tumor.

RESULTS

200 cases, representing a variety of tumor types were analyzed using CancerPlex® v2 Assay. Tumors averaged 6.65 somatic variants (range: 1-18), of which 2.48 variants (range: 0-9) were deemed clinically actionable (93%). Tumor response, or resistance markers were identified for FDA-approved therapies (mean: 1.2; range: 0-5) and clinical trials (mean: 7.98 range: 0-24).

Additional 50 randomly selected clinical cases were characterized using the upgraded CancerPlex® v3 Assay that provides about 2-fold deeper sequence coverage (500x) for the same gene panel. Overall fifty seven alterations were found actionable, those alterations are assigned to 47 cases (94%). None of the samples failed to yield a report.

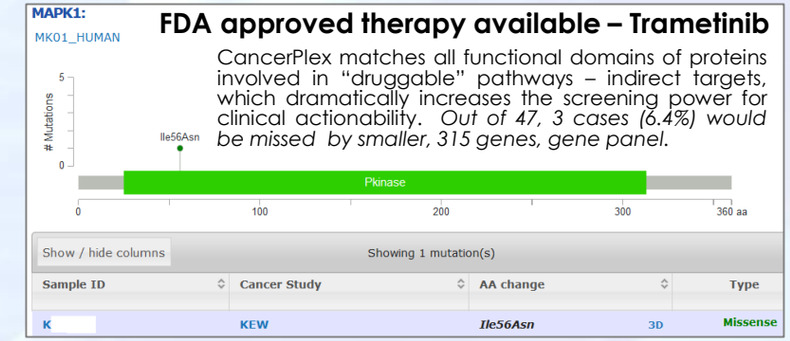




ACTIONABILITY

Our study shows that the actionability rate is not significantly affected by an increase of sequencing coverage. The major reason is the arbitrary cut-off of 10% MAF. The actionability rate could be improved if one were to consider including clonal alterations assigned to only a subset of the entire tumor cell population.

Another factor impacting actionability may be the gene panel size. Published validation studies show that hot-spot analysis (small panels of ~100 cancer genes) misses up to 20% of clinically important mutations in some tumors (Drilon et al., 2015). To test this further, we took a subset of CancerPlex genes to match the content of a previously published smaller panel of 315 genes (Frampton et al., 2015), and compared the actionability. The larger panel provides critically important additions (see **Figures**).



CONCLUSION

Larger NGS panel size increases the likelihood of identifying clinically actionable findings, including FDA-approved therapies missed by NGS panels with fewer genes.

CancerPlex Current Gene List: www.kewgroup.com/sites/default/files/uploads/downloads/kewcancerplexfactsheet.pdf

ABL1	ABL2 (ARG, BACH)	ACVR1B	ACVR2A	AFF1	AKT1	AKT2	AKT3	ALK	AMER1 (FAM123B)	APC	AR	ARAF	ARFRP1 (ARP1)	ARID1A	ARID2	ARNT (HIF-1β)	ASCL4	ASXL1	ATM	ATR	ATRX	AURKA	AURKB	AURKC	AXIN2	AXL (UFO)	BAP1	BARD1	BCL10
BCL11A (ZNF856)	BCL11B	BCL2	BCL2L1 (BCL-X)	BCL2L2 (BCL-W)	BCL6	BCL9	BCOR	BCORL1	BIRC2 (IAP2)	BIRC3 (Aip1)	BLM	BLNK	BMPR1A	BRAF	BRCA1	BRCA2	BRIP1 (FANCJ)	BTK	BUB1B	C11orf30 (EMSY)	CARD11	CASP8	CBFB	CBL	CCND1 (Cyclin D1)	CCND2 (Cyclin-D2)	CCND3 (Cyclin D3)	CCNE1 (Cyclin E1)	CD70 (TNFSF7)
CD79A	CD79B	CDC73 (HRPT1)	CDH1 (E-Cadherin)	CDH11 (Cadherin 11)	CDH2 (N-Cadherin)	CDH5 (VE-Cadherin)	CDK12	CDK4	CDK6	CDK8	CDKN1A	CDKN1B (p27)	CDKN2A (ARF, INK4)	CDKN2B (INK4B)	CDKN2C (INK4C)	CEBPA	CHEK1	CIC	CREBBP	CRKL	CRLF2	CRTC1	CSF1R	CSMD3	CTCF	CTNNA1 (α1-catenin)	CTNNA2 (α2-catenin)	CTNNB1 (β-catenin)	CUX1
CYLD	DAXX	DCC	DDB2	DDIT3 (CHOP)	DDR1	DDR2	DICER1	DNMT3A	DOT1L	DST	EDNRB	EGFR	EML4	EP300 (P300)	EPHA3	EPHA5	EPHA7	EPHB1	EPHB4	EPHB6	ERBB2 (HER2)	ERBB3 (HER3)	ERBB4 (HER4)	ERCC1 (RAD10)	ERCC2 (XPD)	ERCC3 (XPB, RAD25)	ERCC4 (XPF)	ERCC5	ERG
ESR1	ETS1	ETV1	ETV4	EXT1	EXT2	EZH2	FAM46C	FANCA	FANCC	FANCE	FANCF	FANCG	FANCL	FAS (APO-1)	FAT1	FBXW7 (BAF250)	FGF10	FGF14	FGF19	FGF23	FGF3	FGF4	FGF6	FGFR1	FGFR2	FGFR3	FGFR4	FH	FLCN
FLI1	FLT1 (VEGFR1)	FLT3 (STK1)	FLT4 (VEGFR-3)	FN1	FOXA1 (HNF3A)	FOXL2	FOXO1	FOXP1	G6PD	GATA1	GATA2	GATA3	GDNF	GID4	GNA11	GNA13	GNAQ	GNAS	GPC6	GPR124	GRIK3	GRIN2A	GSK3B	HCAR1	HCN1	HGF	HIF1A	HLF	HNF1A
HNF4A	HRAS	HSP90AA1	HSP90AB1	ICK	IDH1	IDH2	IGF1R	IGF2	IGF2R	IKBKB (IKKKB)	IKBKE	IKZF1 (IKAROS)	IL2	IL21R	IL6ST (CD130)	IL7R	INHBA	IRF4	IRS2	ITGA10	ITGB2	ITGB3	JAK1	JAK2	JAK3	JUN	KAT6A (MOZ)	KAT6B (MYST4)	KDM5A (JARID1A)
KDM5C	KDM6A	KDR (VEGFR2)	KEAP1	KIAA1804 (MLK4)	KIT	KLF5	KLF6	KLHL6	MLL (KMT2A)	MLL2 (KMT2D)	KRAS	LAMP1	LCK	LPP	LRP1B	LTF	MAGEA1	MAGI1	MALT1	MAML2	MAP2K1 (MEK1)	MAP2K2 (MEK2)	MAP2K4 (MEK4)	MAP3K1 (MEKK1)	MAP3K7 (TGF1A)	MAPK1 (ERK2)	MAPK8 (JNK1)	MBD1	MCL1
MDM2	MDM4	MED12	MEF2B	MEN1	MET	MITF	MLH1	MMP2	MN1	MPL	MRE11A	MSH2	MSH6	MST1R (RON)	MTOR	MTR	MTRR	MUTYH	MYC	MYCL1	MYCN	MYD88	MYH11	MYH9	NBN	NCOA1	NCOA2	NCOA4	NF1
NF2 (Merlin)	NF2L2 (NRF2)	NFKB1 (NFKB-p105)	NFKB1A	NFKB2 (NFKB)	NIN	NKX2-1 (TTF1)	NLRP1	Notch1	NOTCH2	NOTCH4	NPM1	NRAS	NTRK1	NTRK2	NTRK3	NUMA1	NUP214	NUP93	NUP98	PAK3	PALB2	PARP1	PAX3	PAX5	PAX7	PAX8	PBRM1	PBX1	PDGFRA
PDGFRB	PDK1	PGAP3	PHOX2B	PIK3C2B (PI3K-C2b)	PIK3CA (p110a)	PIK3CB (PIK3C1)	PIK3CD (p110d)	PIK3CG (p110g)	PIK3R1 (GRB1)	PIK3R2 (p85)	PIK3R3 (p55g)	PIM1	PKHD1	PLAG1	PLCG1	PML	PMS1	POT1	PPARG	PPP2R1A	PPP6C	PRDM1 (BLIMP1)	PRKAR1A	PRKDC	PSIP1	PTCH1	PTEN	PTGS2 (COX2)	PTPRD
PTPRT	RAC1	RAF1 (CRAF)	RALGDS	RARA	RASA1	RB1	REL	RET	RHOH	RICTOR	RNASEL	RNF2	RNF43	ROS1	RPS6KA2	RPTOR	RRM1	RUNX1 (AML1)	RUNX1T1 (ETO)	SAMD9	SDHB	SETD2	SF3B1	SGK1	SH2D1A	SLIT2	SMAD2	SMAD4	SMARCA4
SMARCB1	SMO	SOCS1	SOX10	SOX11	SOX2	SOX9	SPEN	SPOP	SRC	STAG2	STAT3	STAT4	STK11 (LKB1)	STK36 (FU)	SUFU	SYK	TAF1	TAF1L	TAL1 (SCL)	TBX22	TCF12	TCF3	TCF7L1	TCF7L2	TCL1A	TET1	TET2	TFE3	TGFBR2
TGM7	TLR4	TNFAIP3 (A20)	TNFRSF14	TNK2 (ACK1)	TOP1	TP53 (P53)	TP63	TPR	TRIM33 (TIF1γ)	TRIP11	TRRAP	TSC1	TSC2	TSHR	TYK2	UBR5	USP9X	VHL	WISP3	WT1	XPC	XPO1 (CRM1)	ZNF384	ZNF521 (EVI-3)					

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