Product Insert

VARIANT*Plex*™ Pan Solid Tumor panel

VARIANT*Plex* Pan Solid Tumor

Description

The VARIANT*Plex* Pan Solid Tumor panel is a balanced pool of gene-specific primer (GSP) oligonucleotides that is optimized for use with VARIANT*Plex* HGC 2.0 reagents and molecular barcode (MBC) adapters to produce targeted NGS libraries. This product insert should be used in conjunction with VARIANT*Plex* HS/HGC protocol for Illumina® (RA-DOC-056).

VARIANT*Plex* Pan Solid Tumor contains **9878** GSPs targeting **185** genes commonly mutated in solid tumors as well as microsatellite instability (**MSI**) and tumor mutational burden (**TMB**) status.

Description	Part number	Storage
VARIANT*Plex* Pan Solid Tumor GSP1, 8 reactions	SA20196081	–20°C ± 10°C
VARIANT*Plex* Pan Solid Tumor GSP2, 8 reactions	SA20196082	
PreSeq™ DNA QC Assay Standard, 32 µL	SA0597	
PreSeq™ DNA QC Assay 10X Primer Mix, 120 µL	SA0598	

Required reagent volumes

Protocol reference	Protocol step	Reagent	Volume per reaction (µL)
A	Ligation Step 2 Elution	5mM NaOH	16
B	First PCR	VARIANT*Plex* Pan Solid Tumor GSP1	24
C	First PCR	10mM Tris-HCl pH 8.0	18
D	First PCR	Purified PCR1 eluate	16
E	Second PCR	VARIANT*Plex* Pan Solid Tumor GSP2	24

Recommended PCR cycling

	Step	Temperature (°C)	Time	Cycles
First PCR reaction	**1**	95	3 min	1
	2	95	30 sec	15
	3	58	10 sec	
	4	60	10 min (100% ramp rate)	
	5	72	3 min	1
	6	4	Hold	1
Second PCR reaction	**1**	95	3 min	1
	2	95	30 sec	20†
	3	58	10 sec	
	4	65	10 min (100% ramp rate)	
	5	72	3 min	1
	6	4	Hold	1

†The number of PCR2 cycles may be decreased if you regularly experience library yields greater than 200 nM.

Recommended reads and multiplexing

VARIANT*Plex* Pan Solid Tumor libraries should be sequenced to a minimum of **25M** reads. Starting read depth recommendations for standard profiling may be adjusted based on user needs.

Archer™ Analysis settings

Sequencing data should be processed using Archer Analysis (v7.2, or greater). The VARIANT*Plex* Pan Solid Tumor panel requires selection of the ***SNV/Indel, Copy Number Variation, Structural Variation, MSI, and TMB*** pipelines, found under the ***DNA*** Input Type**.** The recommended TMB settings in Archer Analysis are *High Threshold for TMB* = 20 and *Low Threshold* for TMB = 5 for VARIANT*Plex* Pan Solid Tumor. As with most Analysis settings, these may be adjusted based on user needs. Selection of the DNA Target Coverage pipeline is also optional and requires a region of interest bed file from supporting. See the Archer Analysis User Guide for more details on setting up your analysis.

Processing of VARIANT*Plex* Pan Solid Tumor libraries requires a one-time upload of a Target Region file (a text file, in GTF format, which directs the software on how to analyze data from the panel). For SNV/Indel detection it is recommended analysis is performed using a Targeted Mutations File. Files can be obtained by contacting archer-tech@idtdna.com

Assay targets

Integrative genomic biomarkers

MSI, TMB

Genes

The following genes in the panel have full coding sequence coverage for SNV/Indel analysis. Coding sequence was determined using NCBI RefSeq transcripts†.

ABL1	*BMPR1A*	*CTNNB1**	*FGF19*	*HNF1A*	*MDM4*	*NOTCH2*
ACVR1	*BRAF**	*DAXX*	*FGFR1*	*HRAS*	*MED12*	*NOTCH3*
AKT1	*BRCA1**	*DDR2*	*FGFR2*	*IDH1*	*MEN1*	*NOTCH4*
AKT2	*BRCA2**	*DDX3X*	*FGFR3*	*IDH2*	*MET**	*NPM1*
AKT3	*BRIP1*	*DICER1*	*FGFR4*	*JAK1*	*MLH1*	*NRAS*
ALK	*CCND1*	*EGFR*	*FH*	*JAK2*	*MPL*	*NTRK1*
APC	*CCND2*	*EIF1AX*	*FLCN*	*JAK3*	*MRE11A*	*NTRK2*
AR	*CCND3*	*EP300*	*FLT1*	*KDM6A*	*MSH2*	*NTRK3*
ARID1A	*CCNE1*	*EPCAM*	*FLT3*	*KDR*	*MSH3*	*PALB2*
ARID1B	*CDH1*	*ERBB2*	*FLT4*	*KEAP1*	*MSH6*	*PBRM1*
ARID2	*CDK12*	*ERBB3*	*FOXA1*	*KIT**	*MTOR*	*PDGFRA**
*ATM**	*CDK4*	*ERBB4*	*FOXL2*	*KLF4*	*MUC16*	*PIK3CA*
ATR	*CDK6*	*ERCC1*	*FUBP1*	*KMT2C*	*MUTYH*	*PIK3CB*
ATRX	*CDKN2A*	*ERCC2*	*GNA11*	*KMT2D (MLL2)*	*MYC*	*PIK3R1**
AURKA	*CDKN2B*	*ESR1*	*GNAQ*	*KRAS*	*MYCN*	*PLCB4*
B2M	*CHD1*	*EZH2*	*GNAS*	*LZTR1*	*NBN*	*PMS2*
BAP1	*CHEK1*	*FANCA*	*H3F3A*	*MAP2K1 (MEK1)*	*NF1*	*POLD1*
BARD1	*CHEK2**	*FANCI*	*H3F3B*	*MAP2K2 (MEK2)*	*NF2*	*POLE*
BCOR	*CIC*	*FANCL*	*HIST1H3B*	*MAP3K1*	*NKX2-1*	*PPP2R1A*

BLM	*CSF1R*	*FBXW7*	*HIST1H3C*	*MDM2*	*NOTCH1**	*PPP2R2A*
PRKD1	*RAD51C*	*RICTOR*	*SETD2*	*SRC*	*TP53*	*VHL*
PTCH1	*RAD51D*	*RNF43*	*SF3B1*	*SRSF2*	*TP63*	*XRCC2*
PTEN	*RAD54L*	*ROS1*	*SMAD2*	*STAG2*	*TRAF7*	*XRCC3*
PTPN11	*RAF1*	*SDHA*	*SMAD4*	*STK11*	*TSC1*	
RAD50	*RB1*	*SDHB*	*SMARCA4*	*SUFU*	*TSC2*	
RAD51	*RET**	*SDHC*	*SMARCB1*	*TERT*	*TSHR*	
RAD51B	*RHOA*	*SDHD*	*SMO*	*TGFBR2*	*U2AF1*	

†Contact adx-tech@idtdna.com for the panel target file to view complete list of targeted regions.
*Indicates that select regions of this gene are enabled for Structural Variant analysis in the GTF.

Genes targeted for CNV

AKT1	*BRIP1*	*ERBB2*	*JAK2*	*PDGFRA*
AKT2	*CCND1*	*ERBB3*	*KDR*	*PIK3CA*
AKT3	*CCND2*	*ERCC1*	*KEAP1*	*PIK3CB*
ALK	*CCND3*	*ERCC2*	*KIT*	*PTEN*
AR	*CCNE1*	*ESR1*	*KRAS*	*RAD51B*
ATM	*CDK12*	*FGF19*	*MDM2*	*RAF1*
AURKA	*CDK4*	*FGFR1*	*MDM4*	*RET*
BARD1	*CDK6*	*FGFR2*	*MET*	*RICTOR*
BRAF	*CHEK1*	*FGFR3*	*MYC*	*STK11*
BRCA1	*CHEK2*	*FGFR4*	*MYCN*	*TERT*
BRCA2	*EGFR*	*FLT3*	*NRAS*	

Please contact archer-tech@idtdna.com to inquire about enabling additional genes for CNV detection.

SNPs and sites targeted for sample tracking

rs560681	rs430046	rs987640	rs10776839	rs12393891
rs740598	rs8078417	rs6444724	rs6530357	chrX:4429309
rs1498553	rs9951171	rs6811238	rs5971553	chrX:11314433
rs10773760	rs576261	rs13182883	rs5953060	chrY:6738552
rs1058083	rs1109037	rs214955	rs6524626	chrY:19490214
rs4530059	rs1523537	rs321198	rs5940270	
rs1821380	rs221956	rs4606077	rs722847	

SNPs may be used in combination to uniquely tag and track samples over time. Contact archer-tech@idtdna.com for further details.

Limitations of use

For research use only. Not for use in diagnostic procedures. Unless otherwise agreed to in writing, IDT does not intend these products to be used in clinical applications and does not warrant their fitness or suitability for any clinical diagnostic use. Purchaser is solely responsible for all decisions regarding the use of these products and any associated regulatory or legal obligations.

Safety data sheets pertaining to this product are available upon request.

Revision History

Document Number	Date	Description of change
RA-DOC-031/REV01	June 2023	Initial release.
RA-DOC-031/REV02	August 2023	Added TMB pipeline information to “Archer Analysis settings” section.
RA-DOC-031/REV03	November 2023	Updated First and Second PCR cycling conditions to include separate anneal and extended steps. Updated branding.