VARIANT*Plex*-HT Core Solid Tumor

Description

The VARIANT*Plex*-HT Core Solid Tumor panel is a balanced pool of gene-specific primer (GSP) oligonucleotides that is optimized for use with VARIANT*Plex*-HT reagents and molecular barcode (MBC) adapters to produce targeted NGS libraries. This product insert should be used in conjunction with VARIANT*Plex*-HT protocol for Illumina® (RA-DOC-058).

VARIANT*Plex*-HT Core Solid Tumor contains **1614** GSPs targeting **60** genes commonly mutated in solid tumors as well as microsatellite instability (MSI).

Description	Part number	Storage
VARIANT*Plex*-HT Core Solid Tumor GSP1, 24 reactions or VARIANT*Plex*-HT Core Solid Tumor GSP1, 96 reactions	SA20122241 or SA20122961	–20°C ± 10°C
VARIANT*Plex*-HT Core Solid Tumor GSP2, 24 reactions or VARIANT*Plex*-HT Core Solid Tumor GSP2, 96 reactions	SA20122242 or SA20122962	

Required reagent volumes

Protocol reference	Protocol step	Reagent	Volume per reaction (µL)
A	Ligation Step 2 Elution	5mM NaOH	20
B	First PCR	VARIANT*Plex*-HT Core Solid Tumor GSP1	8
C	First PCR	10mM Tris-HCl pH 8.0	18
D	First PCR	Purified PCR1 eluate	16
E	Second PCR	VARIANT*Plex*-HT Core Solid Tumor GSP2	8

Recommended PCR cycling

	Step	Temperature (°C)	Time	Cycles
First PCR reaction	**1**	95	3 min	1
	2	95	30 sec	15
	3	60	10 sec	
	4	63	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	60	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*-HT Core Solid Tumor libraries should be sequenced to a minimum of **4.5M** 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 (v7.0, or greater). The VARIANT*Plex*-HT Core Solid Tumor panel requires selection of the ***SNV/Indel, Structural Variation, Copy Number Variation, and MSI*** pipelines, found under the ***DNA*** Input Type (see the Archer Analysis User Guide for more details on setting up your analysis). Selection of the DNA Target Coverage pipeline is optional.

Processing of VARIANT*Plex*-HT Core Solid Tumor libraries requires a one-time upload of the Panel GTF. When performing DNA Target Coverage analysis, users must also select a Region

of Interest BED file. Users may optionally add a Targeted Mutations VCF file for targeted SNV/Indel detection. Files can be obtained by contacting archer-tech@idtdna.com

Assay targets

Gene	Accession	Exon
AKT1	NM_005163	2,3,6,11
ALK	NM_004304	21,22,23,24,25
APC	NM_000038	16
AR	NM_000044	4,5,6,7,8
ATRX	NM_000489	1,2,3,4,5,6,7,8,9,10,11,12,13,14,15,16,17,18,19,20,21,22,23,24,25,26,27,28,29,30,31,32,33,34,35
BRAF	NM_004333	11,15
CDK4	NM_000075	2,3,4,5,6,7,8
CDK6	NM_001259	CNV Only
CDKN2A	NM_000077	2,3
CDKN2A	NM_058197	1
CTNNB1	NM_001904	3
DDR2	NM_006182	8,15,17
EGFR	NM_005228	3,7,12,15,18,19,20,21,22
EGFR	NM_201282	16
EGFR	NM_201283	10
ERBB2	NM_004448	8,10,17,19,20,21,22,24

Gene	Accession	Exon
ERBB3	NM_001005915	3
ERBB3	NM_001982	2,3,7,8,9,18,23,27,28
ERBB4	NM_005235	3,4,6,7,8,9,15,23
ESR1	NM_000125	5,6,7,8
FBXW7	NM_001013415	1
FBXW7	NM_001257069	4
FBXW7	NM_018315	1,2,3,4,5,6,7,8,9,10,11
FGFR1	NM_001174064	3
FGFR1	NM_001174067	2
FGFR1	NM_015850	2,4,5,8,13,14
FGFR2	NM_000141	2,5,7,9,12,13,14,15
FGFR2	NM_022970	8
FGFR3	NM_000142	3,7,9,13,14,16
FOXL2	NM_023067	1 (p.C134)
GNA11	NM_002067	5
GNAQ	NM_002072	4,5
GNAS	NM_000516	6,7,8,9
H3F3A	NM_002107	2
HIST1H3B	NM_003537	1

Gene	Accession	Exon
HRAS	NM_005343	2,3
IDH1	NM_005896	3,4
IDH2	NM_002168	4
JAK2	NM_004972	6,12,14,23
KIT	NM_000222	2,8,9,10,11,12,13,14,15,17,18
KRAS	NM_004985	2,3,4,5
MAP2K1	NM_002755	2,3,6
MAP2K2	NM_030662	2,6
MET	NM_000245	2,11,14,15,16,19,20,21
MTOR	NM_004958	30,39,40,44,45,47,48,50
MYC	NM_002467	1,2,3
NOTCH1	NM_017617	24,25,26,27,28,29,34,c.*7668+371 to c.*7668+378
NOTCH2	NM_024408	24,25,26,27,28,29,34
NOTCH3	NM_000435	24,25,26,27,28,29,33
NOTCH4	NM_004557	26,27,28,29,30
NRAS	NM_002524	2,3,4,5
NTRK1	NM_002529	14,15
NTRK2	NM_006180	18,19
NTRK3	NM_002530	16,17

Gene	Accession	Exon
PDGFRA	NM_006206	7,10,11,12,14,15,16,18,23
PIK3CA	NM_006218	2,3,5,7,8,9,10,14,19,21
POLD1	NM_001308632	14
POLD1	NM_002691	2,3,4,5,6,7,8,9,10,11,12,13,14,16,17,18,19,20,21,22,23,24,25,26,27
POLE	NM_006231	1,2,3,4,5,6,7,8,9,10,11,12,13,14,15,16,17,18,19,20,21,22,23,24,25,26,27,28,29,30,31,32,33,34,35,36,37,38,39,40,41,42,43,44,45,46,47,48,49
PTEN	NM_000314	1,2,3,4,5,6,7,8,9
RAF1	NM_002880	3,7,10,14
RB1	NM_000321	1,2,3,4,5,6,7,8,9,10,11,12,13,14,15,16,17,18,19,20,21,22,23,24,25,26,27
RET	NM_020630	10,11,13,14,15,16
RICTOR	NM_152756	CNV only
ROS1	NM_002944	36,37,38,39,40,41,42
SMAD4	NM_005359	2,3,4,5,6,7,8,9,10,11,12
SMO	NM_005631	1,2,3,4,5,6,7,8,9,10,11,12
TERT	NM_198253	3,6,10
TERT	NM_198253	Promoter (chr5:1295148-1295374)
TP53	NM_000546	1,2,3,4,5,6,7,8,9,10,11
TP53	NM_001276696	10
VHL	NM_000551	1,2,3

Genes targeted for CNV

AKT1	*CDK6*	*FGFR1*	*KRAS*	*PIK3CA*
ALK	*EGFR*	*FGFR2*	*MET*	*PTEN*
AR	*ERBB2*	*FGFR3*	*MYC*	*RAF1*
BRAF	*ERBB3*	*JAK2*	*NRAS*	*RET*
CDK4	*ESR1*	*KIT*	*PDGFRA*	*RICTOR*
				TERT

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-458/REV01	October 2023	Initial release.
RA-DOC-458/REV02	November 2023	Updated First and Second PCR cycling conditions to include separate anneal and extended steps. Added MSI pipeline information to the “Archer Analysis settings” section. Updated branding.