VARIANT*Plex*-HT Myeloid v2

Description

The VARIANT*Plex*-HT Myeloid panel v2 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 Myeloid v2 contains GSPs targeting **92** genes frequently mutated in myeloid malignancies.

Description	Part number	Storage
VARIANT*Plex*-HT Myeloid v2 GSP1, 24 reactions or VARIANT*Plex*-HT Myeloid v2 GSP1, 96 reactions	10029327 or 10029342	–20°C ± 10°C
VARIANT*Plex*-HT Myeloid v2 GSP2, 24 reactions or VARIANT*Plex*-HT Myeloid v2 GSP2, 96 reactions	10029329 or 10029344	

Required reagent volumes

Protocol reference	Protocol Step	Reagent	Volume per reaction (µL)
A	Ligation step 2 elution	5 mM NaOH	20
B	First PCR	VARIANT*Plex*-HT Myeloid v2 GSP1	8
C	First PCR	10 mM Tris-HCl pH 8.0	18
D	First PCR	Purified First PCR eluate	16
E	Second PCR	VARIANT*Plex*-HT Myeloid v2 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	62	5 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	5 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* Myeloid v2 libraries should be sequenced to the minimum depth required for the desired variant allele frequency (VAF).

Recommended Reads	Desired VAF
1.5M	5% or greater
10M	1%

Archer™ Analysis settings

Sequencing data should be processed using Archer Analysis (v7.0, or greater). The VARIANT*Plex*-HT Myeloid v2 panel requires selection of the ***SNV/Indel, Copy Number Variation (CNV), CNV 2.0, and Structural Variation*** 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 Myeloid v2 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
ABL1	NM_005157	4,5,6,7,8,9,10
ANKRD26	NM_014915	1 (c.-113-c.-134)
ASXL1	NM_015338.5	1,2,3,4,5,6,7,8,9,10,11,12,13
ASXL1	NM_001164603.1	5
ATRX	NM_000489	8,9,10,11,17,18,19,20,21,22,23,24,25,26,27,28,29,30,31,32
BCOR	NM_017745	2,3,4,5,6,7,9,10,11,12,13,14,15
BCOR	NM_001123385	8
BCORL1	NM_021946	1,2,3,4,5,6,7,8,9,10,11,12
BRAF	NM_004333	3,10,11,12,13,15
BRCC3	NM_024332	1,2,3,4,5,6,7,8,9,10,11
BTK	NM_000061	15
CALR	NM_004343	8,9
CBL	NM_005188	2,3,4,5,7,8,9,16
CBLB	NM_170662	3,9,10
CBLC	NM_012116	9,10
CCND2	NM_001759	5

Gene	Accession	Exon
CDKN2A	NM_058197	1
CDKN2A	NM_058195	1
CDKN2A	NM_000077	2,3
CDKN2A	NM_001195132	3
CEBPA	NM_004364	1
CREBBP	NM_004380	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
CTCF	NM_006565	3,4,5,6,7,8,9,10,11,12
CSF3R	NM_156039	17
CSF3R	NM_172313	10,18
CSF3R	NM_000760	14,15,16
CUX1	NM_001202543	15,16,17,18,19,20,21,22,23,24
CUX1	NM_001913	1,2,3,4,5,6,7,8,9,10,11,12,13,14,15,16,17,18,19,20,21,22,23
CUX1	NM_181552	1
CXCR4	NM_003467	1,2
DCK	NM_000788	2,3
DDX41	NM_016222	1,2,3,4,5,6,7,8,9,10,11,12,13,14,15,16,17
DHX15	NM_001358	3
DNMT3A	NM_022552	2,3,5,6,7,8,9,10,11,12,13,14,15,16,17,18,19,20,21,22,23
DNMT3A	NM_153759	1,2
DNMT3A	NM_175630	4
EED	NM_003797	1,2,3,4,5,6,7,8,9,10,12
EED	NM_152991	11
EED	NM_001308007	10
ETNK1	NM_018638	3

Gene	Accession	Exon
ETV6	NM_001987	1,2,3,4,5,6,7,8
EZH2	NM_004456	2,3,4,5,6,7,8,9,10,11,12,13,14,15,16,17,18,19,20
FBXW7	NM_018315	1,2,3,4,5,6,7,8,9,10,11
FLT3	NM_004119	8,9,10,11,12,13,14,15,16,17,19,20,21
GATA1	NM_002049	2,3
GATA2	NM_032638	2,3,4,5,6
GNAS	NM_000516	8,9,10,11
GNB1	NM_002074	3,4,5,6,7,8,9,10,11
HRAS	NM_005343	2,3,4
IDH1	NM_005896	3,4
IDH2	NM_002168	4,6
IKZF1	NM_001291845	4
IKZF1	NM_001291846	5
IKZF1	NM_006060	2,3,4,5,6,7,8
IL7R	NM_002185	5,6
JAK1	NM_002227	3,4,6,8,10,11,13,14,15,16,17,19,20
JAK2	NM_004972	12,13,14,15,16,19,20,21,22,23,24,25
JAK3	NM_000215	3,5,11,13,15,16,18,19,20,21
KDM6A	NM_021140	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
KDM6A	NM_001291415	14
KIT	NM_000222	1,2,5,8,9,10,11,12,13,14,15,17,18
KMT2A	NM_005933	1,2,3,4,5,6,7,8,9,10,11,12,13,15,16,17,18,19,20,21,22,23,24,25,26,27,28,29,30, 31,32,33,34,35,36
KMT2A	NM_001197104	14
KMT2D	NM_003482	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, 31,32,33,34,35,36,37,38,39,40,41,42,43,44,45,46,47,48,49,50,51,52,53,54,55

Gene	Accession	Exon
KRAS	NM_004985	2,3,4,5
KRAS	NM_033360	5
LUC7L2	NM_016019	1,2,3,4,5,6,7,8,9,10
LUC7L2	NM_001244585	2
MAP2K1	NM_002755	2,3
MPL	NM_005373	10,12
MYC	NM_002467	1,2,3
MYD88	NM_002468	4,5
MYD88	NM_001172567	3
NF1	NM_000267	1,2,3,4,5,6,7,8,9,10,11,12,13,14,16,17,18,19,20,21,22,23,24,25,26,27,28,29,30,31,32,33,34,35,36,37,38,39,40,41,42,43,44,45,46,47,48,49,50,51,52,53,54,55,56,57
NF1	NM_001128147	15
NF1	NM_001042492	31
NOTCH1	NM_017617	26,27,28,31,32,33, 34,c.*370 to c.*380
NPM1	NM_002520	11
NRAS	NM_002524	2,3,4,5
NUDT15	NM_018283	p.M1T, p.M1I ,p.del17_18GV,p.V18I,p.C28fs,p.L30V,p.R34T,p.K35E ,p.G47R,p.F52L, p.N74fs,p.E115fs,p.E118X,p.P129R,p.R139C,p.R139H,p.L156Q
PAX5	NM_016734	1,2,3,4,5,6,7,8,9
PAX5	NM_001280549	8
PDGFRA	NM_006206	12,14,15,18
PHF6	NM_032335	2,3,4,5,6,7,8
PHF6	NM_001015877	10
PHF6	NM_032458	9
PPM1D	NM_003620	6

Gene	Accession	Exon
PRPF8	NM_006445	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
PTEN	NM_000314	1,2,3,4,5,6,7,8,9
PTPN11	NM_002834	3,4,7,8,12,13
PTPN11	NM_080601	11
RAD21	NM_006265	2,3,4,5,6,7,8,9,10,11,12,13,14
RBBP6	NM_006910	p.1444,p.1451,p.1569,p.1654,p.1673
RUNX1	NM_001754	2,3,5,6,7,8,9
RUNX1	NM_001122607	1,5
SAMD9	NM_017654	3
SAMD9L	NM_152703	5
SETBP1	NM_015559	4,p.799-p.950
SF3B1	NM_012433	13,14,15,16,17,18,19,20,21
SH2B3	NM_005475	2,3,4,5,6,7,8
SLC29A1	NM_001078175	4,13
SMC1A	NM_006306	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
SMC1A	NM_001281463	2
SMC3	NM_005445	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
SRSF2	NM_003016	1,2
STAG2	NM_006603	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
STAG2	NM_001042749	32
STAT3	NM_003150	20
STAT3	NM_139276	21
STAT5B	NM_012448	15,16
TET2	NM_001127208	4,5,6,7,8,9,10,11

Gene	Accession	Exon
TET2	NM_017628	3
TPMT	NM_000367	p.Y240C,p.Y240S,p.K238E,p.H227Q,p.R226Q,p.C216X,p.R215H,p.C212R, p.F208L,p.I204T,p.V199I,p.Y180F,p.Q179H,p.S172P,p.A167G,p.Y166C, p.R163H,p.R163P,p.R163C,p.A154T,p.G144R,p.C132Y,p.S125L,p.K122T, p.K119T,p.G117R,p.E114K,p.Y107D,p.G88S,p.R82W,p.A80P,p.W78C,p.A73V, p.G71R,p.L69V,p.F67S,p.L49S,p.Q42E,p.G36S,p.K32KfsX58, p.E28V,p.M1T,p.M1V
TP53	NM_000546	1,2,3,4,5,6,7,8,9,10,11
TP53	NM_001276696	10
TP53	NM_001276695	10
UBA1	NM_003334	3
UBTF	NM_014233	2,3,4,5,6,7,8,9,10,11,12,13,14,15,16,17,18,19,20,21
U2AF1	NM_006758	2,6,7
U2AF1	NM_001025204	6
U2AF2	NM_007279	1,2,3,4,5,6,7,8,9,10,11,12
WT1	NM_000378	1,2,3,4,5,6,7,9
WT1	NM_024426	5
WT1	NM_001198552	8
XPO1	NM_003400	15,16,18
ZRSR2	NM_005089	1,2,3,4,5,6,7,8,9,10,11

Genes targeted for CNV

ASXL1	*CDKN2A*	*FLT3*	*MYC*	*RUNX1*	*U2AF2*
BCOR	*CUX1*	*IKZF1*	*NF1*	*TET2*	*WT1*
CBL	*ETV6*	*KDM6A*	*RAD21*	*TP53*	*ZRSR2*
CDC25C	*EZH2*	*LUC7L2*	*RPS14*	*U2AF1*	

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-616/REV01	May 2025	Initial release.