

VARIANTPlex-HT Myeloid v2

Description

The VARIANTPlex-HT Myeloid v2 panel is a balanced pool of gene-specific primer (GSP) oligonucleotides that is optimized for use with VARIANTPlex-HT reagents and molecular barcode (MBC) adapters to produce targeted NGS libraries. This product insert should be used in conjunction with VARIANTPlex-HT protocol for Illumina® (RA-DOC-058).

VARIANTPlex-HT Myeloid v2 contains **2612** GSPs targeting **92** genes frequently mutated in myeloid malignancies.

Description	Part number	Storage
VARIANTPlex-HT Myeloid v2 GSP1, 24 reactions or VARIANTPlex-HT Myeloid v2 GSP1, 96 reactions	10029327 or 10029342	-20°C ± 10°C
VARIANTPlex-HT Myeloid v2 GSP2, 24 reactions or VARIANTPlex-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	VARIANTPlex-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	VARIANTPlex-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

VARIANTPlex 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 VARIANTPlex-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 VARIANTPlex-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
<i>ABL1</i>	NM_005157	4,5,6,7,8,9,10
<i>ANKRD26</i>	NM_014915	1 (c.-113-c.-134)
<i>ASXL1</i>	NM_015338.5	1,2,3,4,5,6,7,8,9,10,11,12,13
<i>ASXL1</i>	NM_001164603.1	5
<i>ATRX</i>	NM_000489	8,9,10,11,17,18,19,20,21,22,23,24,25,26,27,28,29,30,31,32
<i>BCOR</i>	NM_017745	2,3,4,5,6,7,9,10,11,12,13,14,15
<i>BCOR</i>	NM_001123385	8
<i>BCORL1</i>	NM_021946	1,2,3,4,5,6,7,8,9,10,11,12
<i>BRAF</i>	NM_004333	3,10,11,12,13,15
<i>BRCC3</i>	NM_024332	1,2,3,4,5,6,7,8,9,10,11
<i>BTK</i>	NM_000061	15
<i>CALR</i>	NM_004343	8,9
<i>CBL</i>	NM_005188	2,3,4,5,7,8,9,16
<i>CBLB</i>	NM_170662	3,9,10
<i>CBLC</i>	NM_012116	9,10
<i>CCND2</i>	NM_001759	5

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

Gene	Accession	Exon
<i>ETV6</i>	NM_001987	1,2,3,4,5,6,7,8
<i>EZH2</i>	NM_004456	2,3,4,5,6,7,8,9,10,11,12,13,14,15,16,17,18,19,20
<i>FBXW7</i>	NM_018315	1,2,3,4,5,6,7,8,9,10,11
<i>FLT3</i>	NM_004119	8,9,10,11,12,13,14,15,16,17,19,20,21
<i>GATA1</i>	NM_002049	2,3
<i>GATA2</i>	NM_032638	2,3,4,5,6
<i>GNAS</i>	NM_000516	8,9,10,11
<i>GNB1</i>	NM_002074	3,4,5,6,7,8,9,10,11
<i>HRAS</i>	NM_005343	2,3,4
<i>IDH1</i>	NM_005896	3,4
<i>IDH2</i>	NM_002168	4,6
<i>IKZF1</i>	NM_001291845	4
<i>IKZF1</i>	NM_001291846	5
<i>IKZF1</i>	NM_006060	2,3,4,5,6,7,8
<i>IL7R</i>	NM_002185	5,6
<i>JAK1</i>	NM_002227	3,4,6,8,10,11,13,14,15,16,17,19,20
<i>JAK2</i>	NM_004972	12,13,14,15,16,19,20,21,22,23,24,25
<i>JAK3</i>	NM_000215	3,5,11,13,15,16,18,19,20,21
<i>KDM6A</i>	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
<i>KDM6A</i>	NM_001291415	14
<i>KIT</i>	NM_000222	1,2,5,8,9,10,11,12,13,14,15,17,18
<i>KMT2A</i>	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
<i>KMT2A</i>	NM_001197104	14
<i>KMT2D</i>	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
<i>KRAS</i>	NM_004985	2,3,4,5
<i>KRAS</i>	NM_033360	5
<i>LUC7L2</i>	NM_016019	1,2,3,4,5,6,7,8,9,10
<i>LUC7L2</i>	NM_001244585	2
<i>MAP2K1</i>	NM_002755	2,3
<i>MPL</i>	NM_005373	10,12
<i>MYC</i>	NM_002467	1,2,3
<i>MYD88</i>	NM_002468	4,5
<i>MYD88</i>	NM_001172567	3
<i>NF1</i>	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
<i>NF1</i>	NM_001128147	15
<i>NF1</i>	NM_001042492	31
<i>NOTCH1</i>	NM_017617	26,27,28,31,32,33, 34,c.*370 to c.*380
<i>NPM1</i>	NM_002520	11
<i>NRAS</i>	NM_002524	2,3,4,5
<i>NUDT15</i>	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
<i>PAX5</i>	NM_016734	1,2,3,4,5,6,7,8,9
<i>PAX5</i>	NM_001280549	8
<i>PDGFRA</i>	NM_006206	12,14,15,18
<i>PHF6</i>	NM_032335	2,3,4,5,6,7,8
<i>PHF6</i>	NM_001015877	10
<i>PHF6</i>	NM_032458	9
<i>PPM1D</i>	NM_003620	6

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

Gene	Accession	Exon
<i>TET2</i>	NM_017628	3
<i>TPMT</i>	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
<i>TP53</i>	NM_000546	1,2,3,4,5,6,7,8,9,10,11
<i>TP53</i>	NM_001276696	10
<i>TP53</i>	NM_001276695	10
<i>UBA1</i>	NM_003334	3
<i>UBTF</i>	NM_014233	2,3,4,5,6,7,8,9,10,11,12,13,14,15,16,17,18,19,20,21
<i>U2AF1</i>	NM_006758	2,6,7
<i>U2AF1</i>	NM_001025204	6
<i>U2AF2</i>	NM_007279	1,2,3,4,5,6,7,8,9,10,11,12
<i>WT1</i>	NM_000378	1,2,3,4,5,6,7,9
<i>WT1</i>	NM_024426	5
<i>WT1</i>	NM_001198552	8
<i>XPO1</i>	NM_003400	15,16,18
<i>ZRSR2</i>	NM_005089	1,2,3,4,5,6,7,8,9,10,11

Genes targeted for CNV

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

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

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Revision History

Document Number	Date	Description of change
RA-DOC-616/REV01	May 2025	Initial release.
RA-DOC-616/REV02	Aug 2025	Added GSP2 counts