FUSION*Plex* ALL Panel

Description

The FUSION*Plex* ALL panel is a balanced pool of gene-specific primer (GSP) oligonucleotides that is optimized for use with FUSION*Plex* reagents and molecular barcode (MBC) adapters to produce targeted NGS libraries. This product insert should be used in conjunction with FUSION*Plex* protocol for Illumina® (RA-DOC-047) or FUSION*Plex* protocol for Ion Torrent™ (RA-DOC-048).

FUSION*Plex* ALL contains **506** GSPs targeting **81** genes commonly mutated in Acute Lymphoblastic Leukemia.

Description	Part number	Storage
FUSION*Plex* ALL GSP1, 8 reactions	SA0074-8-1	–20°C ± 10°C
FUSION*Plex* ALL GSP2, 8 reactions	SA0074-8-2	
10X VCP Primer Mix	SA0126	

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	65	5 min (100% ramp rate)	
	5	72	3 min	1
	6	4	Hold	1

Recommended PCR cycling *(cont.)*

	Step	Temperature (°C)	Time	Cycles
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

FUSION*Plex* ALL libraries should be sequenced to a minimum of **1.5M 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 FUSION*Plex* ALL panel requires selection of the ***Fusion*** and (optional) ***SNV/Indel*** pipeline(s), found under the ***RNA*** Input Type (see the Archer Analysis User Guide for more details on setting up your analysis).

Processing of FUSION*Plex* ALL libraries requires a one-time upload of the Panel GTF. Files can be obtained by contacting archer-tech@idtdna.com.

Assay targets

Gene	Accession	Exon	Variant Type	Description*
ABL1	NM_005157	N/A	Mutation	Y253-E255, V299, T315-F317, M351-F359
ABL1	NM_005157	1, 2, 3, 4, 5	Fusion	5'
ABL1	NM_005157	N/A	Expression imbalance	N/A
ABL2	NM_005158	2, 3, 4, 5, 6, 7, 8	Fusion	5'

Gene	Accession	Exon	Variant Type	Description*
AICDA	NM_020661	N/A	Expression	N/A
BCL11B	NM_138576	3, 4	Fusion	5'
BCL11B	NM_138576	2, 3	Fusion	3'
BCL2	NM_000633	N/A	Mutation	F104
BCL2	NM_000633	N/A	Expression	N/A
BCL2	NM_000633	3	Fusion	3'
BCL2	NM_000633	2	Fusion	5'
BCL6	NM_001706	2, 3	Fusion	5'
BCL6	NM_001706	N/A	Expression	N/A
BCR	NM_004327	1, 2, 3, 8, 12, 13, 14, 15, 16	Fusion	3'
BLNK	NM_013314	N/A	Expression	N/A
BRAF	NM_004333	N/A	Mutation	V600
CD274	NM_014143	N/A	Expression	N/A
CHD1	NM_001270	1,2	Fusion	5'
CREBBP	NM_004380	N/A	Mutation	P1053, C1240, R1446, S1680-L1681
CREBBP	NM_004380	2, 3, 4, 5, 6	Fusion	5'
CRLF2	NM_022148	N/A	Mutation	F232
CRLF2	NM_022148	1	Fusion	5'
CRLF2	NM_022148	N/A	Expression	N/A

Gene	Accession	Exon	Variant Type	Description*
CSF1R	NM_005211	9, 10, 11, 12, 13, 14	Fusion	5'
CTLA4	NM_005214	N/A	Expression	N/A
DNM2	NM_004945	N/A	Mutation	V649, L789
DNTT	NM_004088	N/A	Expression	N/A
EBF1	NM_024007	10, 11, 12, 13, 14, 15	Fusion	3'
EPOR	NM_000121	7, 8	Fusion	3'
ETV6	NM_001987	N/A	Mutation	Y104-R105
ETV6	NM_001987	1, 2, 3, 4, 5, 6	Fusion	3'
ETV6	NM_001987	2, 3, 4, 5, 6	Fusion	5'
EZH2	NM_004456	N/A	Mutation	Y646, R690
FBXW7	NM_033632	N/A	Mutation	R224-T226, R338-I347, T385, R465, R479, R505
FGFR1	NM_023110	12, 17	Fusion	3'
FGFR1	NM_023110	2, 3, 4, 5, 6, 7, 8, 9, 10, 11, 17	Fusion	5'
FLT3	NM_004119	N/A	Mutation	F590-N609, D835-S838
FLT3	NM_004119	N/A	Expression	N/A
HOXA10	NR_037939	N/A	Expression	N/A
HOXA10	NM_018951	N/A	Expression	N/A
HOXA9	NM_152739	N/A	Expression	N/A
IDH1	NM_005896	N/A	Mutation	R132

Gene	Accession	Exon	Variant Type	Description*
IDH2	NM_002168	N/A	Mutation	R140, R172
IKZF1	NM_006060	1, 2, 3	Exon skipping	3'
IKZF1	NM_006060	7, 8	Exon skipping	5'
IKZF2	NM_016260	3, 4	Fusion	3'
IKZF3	NM_012481	2, 3, 4, 5, 6, 7	Fusion	3'
IKZF3	NM_012481	N/A	Mutation	L162
IL7R	NM_002185	N/A	Mutation	S185, P240-S246
IRF4	NM_002460	N/A	Expression	N/A
IRF8	NM_002163	N/A	Expression	N/A
JAK1	NM_002227	N/A	Mutation	V658, S703, R724
JAK2	NM_004972	N/A	Mutation	F537-F547, V617-C618, L681-R683, L855, V863, A880, V911, M929-R938, I960, R980-E985, D994
JAK2	NM_004972	6, 7, 8, 9, 10, 11, 12, 13, 15, 16, 17, 18, 19, 20	Fusion	5'
JAK2	NM_004972	9, 10, 11, 12	Fusion	3'
JAK3	NM_000215	N/A	Mutation	M511, A572-A573, R657, S789
KDM6A	NM_021140	N/A	Mutation	V1113
KLF2	NM_016270	2, 3	Fusion	5'
KMT2A	NM_005933	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	Fusion	3'

Gene	Accession	Exon	Variant Type	Description*
KRAS	NM_004985	N/A	Mutation	G12-G13, Q61, A146
LMO1	NM_002315	N/A	Expression	N/A
LYL1	NM_005583	N/A	Expression	N/A
MLLT4	NM_001040000	2	Fusion	5'
MPL	NM_005373	N/A	Mutation	S505, W515
MYC	NM_002467	N/A	Expression	N/A
MYC	NM_002467	1, 2	Fusion	5'
NF1	NM_000267	14	Fusion	3'
NF1	NM_000267	36	Fusion	5'
NOTCH1	NM_017617	N/A	Mutation	L1574, V1578, L1585, F1592-L1593, R1598-L1600, L1678-I1680, P2514-E2515, P2525
NOTCH1	NM_017617	24, 25, 26, 27, 28, 29	Fusion	5'
NOTCH1	NM_017617	24	Fusion	3'
NOTCH1	NM_017617	34	Exon skipping	N/A
NRAS	NM_002524	N/A	Mutation	G12-G13, G60-Q61
NT5C2	NM_012229	N/A	Mutation	R238, R367
NTRK3	NM_002530	N/A	Expression imbalance	N/A
NTRK3	NM_002530	13, 14, 15	Fusion	5'
NTRK3	NM_001007156	15	Fusion	5'
NUP214	NM_005085	17, 18, 19	Fusion	5'

Gene	Accession	Exon	Variant Type	Description*
NUP98	NM_016320	8, 9, 10, 11, 12, 13, 14, 15, 16, 17	Fusion	3'
NUP98	NM_016320	12, 13	Fusion	5'
P2RY8	NM_178129	1	Fusion	3'
PAG1	NM_018440	2	Fusion	5'
PAX5	NM_016734	N/A	Mutation	P80
PAX5	NM_016734	1, 4, 5, 6, 7, 8	Fusion	3'
PAX5	NM_016734	6, 7, 8	Fusion	5'
PBX1	NM_002585	1, 2, 3, 4, 5, 6, 7, 8, 9	Fusion	5'
PDCD1	NM_005018	N/A	Expression	N/A
PDCD1LG2	NM_025239	N/A	Expression	N/A
PDCD1LG2	NM_025239	1, 2, 3	Fusion	5'
PDCD1LG2	NM_025239	5, 6	Fusion	3'
PDGFRA	NM_006206	N/A	Mutation	T674
PDGFRA	NM_006206	9, 10, 11, 12, 13, 14	Fusion	5'
PDGFRB	NM_002609	8, 9, 10, 11, 12, 13, 14	Fusion	5'
PICALM	NM_007166	16, 17, 18, 19	Fusion	3'
PTK2B	NM_173176	2, 3, 4, 5, 6, 7, 8	Fusion	5'
PTPN11	NM_002834	N/A	Mutation	G60-D61, E69-T73, E76, S502-G503
RAG1	NM_000448	N/A	Expression	N/A

Gene	Accession	Exon	Variant Type	Description*
RAG2	NM_000536	N/A	Expression	N/A
RUNX1	NM_001754	2, 3, 4, 5, 6, 7, 8	Fusion	3'
RUNX1	NM_001754	5, 6, 7, 8, 9	Fusion	5'
RUNX1	NM_001754	N/A	Expression imbalance	N/A
SEMA6A	NM_020796	1, 2	Fusion	3'
SETD2	NM_014159	1, 2, 3, 4, 5, 6, 7, 8, 9, 10, 11, 12	Fusion	3'
SH2B3	NM_005475	N/A	Mutation	E208, D231-D234
SOX11	NM_003108	N/A	Expression	N/A
STAT3	NM_003150	N/A	Mutation	Y640-N647, D661
STAT5B	NM_012448	N/A	Mutation	N642
STIL	NM_003035	1, 2	Fusion	3'
TAL1	NM_003189	N/A	Expression	N/A
TAL1	NM_003189	2, 4	Fusion	5'
TAL1	NM_001290404	2, 3	Fusion	5'
TCF3	NM_003200	11, 12, 13, 14, 15, 16, 17, 18	Fusion	3'
TLX1	NM_005521	N/A	Expression	N/A
TLX3	NM_021025	N/A	Expression	N/A
TYK2	NM_003331	16, 18	Fusion	5'
TYK2	NM_003331	N/A	Mutation	W327

Gene	Accession	Exon	Variant Type	Description*
WT1	NM_000378	N/A	Mutation	Q301-T303, G352-Q354, V359-T365, Q445-F448
WT1	NM_000378	N/A	Expression	N/A
ZCCHC7	NM_032226	1, 2	Fusion	3'
ZCCHC7	NM_032226	2, 3, 4	Fusion	5'

*The mutations listed under the Description column are targeted by the assay design. Version 6.2 and earlier of Archer Analysis may not support RNA SNV/Indel variant calling at exon junctions depending on the sequence context (SNVs ≤5bp, indels ≤30bp). *De Novo* RNA SNV/Indel and Internal Tandem Duplication mutation detection are not supported on the Ion Torrent Sequencing System.

Note: Fusions involving *BCR* and *TCR* loci, including *IGH*, *IGL*, and *IGK*, are targeted for expression and may not be explicitly called as a fusion because these often do not result in chimeric transcripts. For the "Expression" assay type, unique molecules originating from probes across these genes can be counted and normalized to target control genes to enable relative expression level detection. Results are visualized in Archer Analysis.

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-008/REV01	June 2023	Initial release
RA-DOC-008/REV02	November 2023	Updated First and Second PCR cycling conditions to include separate anneal and extended steps. Updated branding.