

VARIANT*Plex* CFTR v2

Description

The VARIANT*Plex* CFTR v2 panel is a balanced pool of gene-specific primer (GSP) oligonucleotides that is optimized for use with VARIANT*Plex* reagents and molecular barcode (MBC) adapters to produce targeted NGS libraries. This product insert should be used in conjunction with VARIANT*Plex* standard protocol (RA-DOC-057).

VARIANT*Plex* CFTR v2 contains **208** GSPs with coverage of all exons of the Cystic Fibrosis Conductance Regulator (CFTR) gene, including 5' and 3' untranslated regions (UTRs) and select intronic variants.

Description	Part number	Storage
VARIANT <i>Plex</i> CFTR v2 GSP1, 8 reactions	SA24000081	-20°C ± 10°C
VARIANT <i>Plex</i> CFTR v2 GSP2, 8 reactions	SA24000082	

Required reagent volumes

Protocol reference	Protocol step	Reagent	Volume per reaction (µL)
A	Cleanup after Adapter Ligation	10mM Tris-HCl pH 8.0	20
B	First PCR	VARIANT <i>Plex</i> CFTR v2 GSP1	2
C	First PCR	Purified DNA	18
D	Cleanup after First PCR	10mM Tris-HCl pH 8.0	20
E	Cleanup after First PCR	Purified DNA	18
F	Second PCR	VARIANT <i>Plex</i> CFTR v2 GSP2	2

Recommended PCR cycling

	Step	Temperature (°C)	Time	Cycles
First PCR reaction	1	95	3 min	1
	2	95	30 sec	
	3	60	10 sec	14
	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	
	3	60	10 sec	22†
	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* CFTR v2 libraries should be sequenced to a minimum of **500,000 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 Version, or greater). The VARIANT*Plex* CFTR v2 panel requires selection of the **SNV/Indel and Structural Variation** pipeline, 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* CFTR 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

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.

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Revision History

Document Number	Date	Description of change
RA-DOC-466/REV01	December 2023	Initial release.
RA-DOC-466/REV02	May 2024	Added "Required reagent volumes" table.