

CleanPlex® for MGI SARS-CoV-2 Research and Surveillance Panel

Targeted Sequencing NGS Panels to Support SARS-CoV-2 Research and Surveillance

Highlights

- **Complete Coverage and Comprehensive Data**
Sequence the entire SARS-CoV-2 genome with over 99% coverage
- **Ultra-sensitive Detection**
Detect down to one copy per reaction for degraded or limited sample input
- **Fast, Streamlined Workflow**
Generate libraries for DNBSEQ™ in just 5 hours using a simple, three-step protocol, compatible with automation.
- **Superb Performance**
Prepare high-quality NGS libraries with excellent coverage uniformity and on-target performance to enable efficient use of sequencing reads and reduce costs

Real Time RT-PCR and antibody-based methods are the main tools for detecting infectious agents, however, such methods can only focus on a limited number of targets and can at times suffer from low assay sensitivity and false negatives results. These methods can only provide positive or negative results. The CleanPlex technology is an assay platform for ultra-sensitive and highly-multiplexed PCR-based targeted sequencing tests. This technology provides an easy-to-use, fast, and comprehensive solution for detection, variant identification, and mutation analysis of infectious pathogens all via a quick and easy workflow.

The CleanPlex® for MGI SARS-CoV-2 Research and Surveillance Panel was expertly designed using a proprietary design pipeline to cover the entire genome. For additional flexibility, this panel is available for sequencing on Illumina® and Ion Torrent simply by applying the respective indexing primers in the workflow. The NGS panel not only allows high sensitivity detection and confirmation of questionable qPCR results, but also enables mutation analysis, tracking, surveillance, and informed infection control through comprehensive sequence information generated.

CleanPlex for MGI SARS-CoV-2 Research and Surveillance Panel Specifications

Parameter	Specification
Enrichment Method	Multiplex PCR
Platform	DNBSEQ™
Variant Identification	Yes
Cumulative Target Size	29,903 bp
Number of Amplicons	343
Amplicon Size	116 - 196 bp, Median 149bp
Number of Primer Pools	2
Sample Input Requirement	5-11 µL of extracted total RNA or ~50ng purified total RNA
Sample Types	Sputum, nasopharyngeal and oropharyngeal swabs and aspirate, tissue samples, and other methods for viral RNA sampling.
Total Assay Time	5.5 hours
Hand on Time	Less than 1 hour
Design Coverage	Complete coverage (except 92 bp at the ends of the genome)
Amplicon Coverage (≥50x)	>95% with 300 copies viral input at 0.2M PE Reads per sample
On-Target Aligned Reads	>98% with 300 copies viral input at 0.2M PE Reads per sample
Total Reads per sample	0.2 to 0.3 M PE per sample with 2 x 150 PE reads

CleanPlex Streamlined Targeted Sequencing Workflow

CleanPlex for MGI SARS-CoV-2 Panels offer a simple and streamlined workflow. Starting from purified RNA, the protocol can be completed to generate target-enriched NGS libraries in just 5 hours, with less than 1hr of hands-on time, using a three-step workflow with minimal tube-to-tube transfers.



CleanPlex Target Enrichment and Library Preparation
5 hours of total assay time, with less than 1 hour of hands-on time.

FOR RESEARCH USE ONLY. NOT FOR USE IN DIAGNOSTIC PROCEDURES.

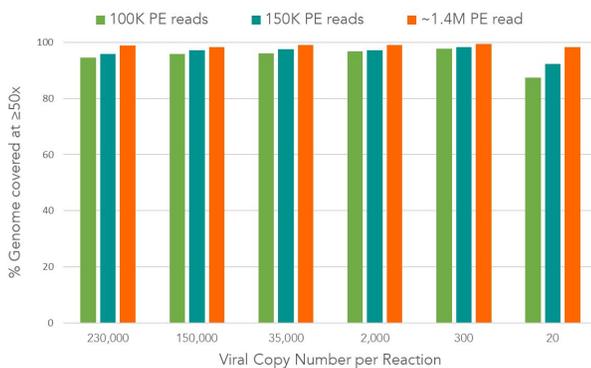
1 | PS5004-03

High Performance Translates to Cost-Effective Sequencing and Confident Analysis

Specifications	20 copies	300 copies
% Uniformity (0.2x Mean)	87%	97%
% Mapping Rate	84%	98%
% On-Target Rate	93%	99%

CleanPlex Panels exhibit highly uniform coverage even with low template input and ultra high multiplexing. Without the need for deep sequencing to capture all targets, high sequencing performance allows for more cost-effective sequencing with more samples pooled per chip for higher throughput.

Genomic Coverage (min 50x) vs Viral Copy Input at Various Sequencing Depths



Using nasopharyngeal swab samples from COVID-19 patients spanning 4 to 40,000 copies/μL (5.5 μL input/reaction) libraries were prepared and sequenced at ~0.7 million cluster reads per sample. The plot above shows *in silico* down sampling data indicating high coverage even at low reads and copy numbers. At the original sequencing depth, >98% of the amplicons are covered at >50X, more than sufficient for mutation monitoring and phylogenetic analysis. For strain confirmation of samples with as little as 4 copies/uL of viral RNA, just 0.1M PE reads is sufficient.

Base Position	Reference base	Alternative Base	Call Frequency
19065	T	C	99.4%
22303	T	G	99.0%
26144	G	T	99.3%
29749	ACGATCGAGTG	A	99.5%

CleanPlex Libraries were created using Twist's control synthetic SARS-CoV-2 MT007544 strain RNA, and variant calling was performed against reference genome NC_045512.2 (MN908947). As shown in the table above, all four expected variants (3 SNVs and 1 deletion) were confidently detected with 99% calling frequency.

Sequencing suggestions for SARS-CoV-2 Panel on DNBSEQ

CleanPlex for MGI panels are specifically designed for 100bp PE reads on the DNBSEQ platforms. Suggested average read per amplicon is 75x or 50,000 cluster reads per sample with 2-pool workflow. More reads might be needed to achieve deeper coverage per amplicon and higher genome coverage for low viral copy number detection or low percentage variant calling.

MGISP-960 Automated Liquid Handling System Compatible	
User-defined	24-position board supporting multiple combinations
Efficient & Accurate	96-channel pipettor with a robotic gripper to ensure high precision and reproducibility.
Hands-Free	Complete CleanPlex SARS-CoV-2 Panel workflow fully integrated.

Ordering Information

The CleanPlex for MGI SARS-CoV-2 Research and Surveillance Panel contains panel mPCR Primers and CleanPlex Targeted Library Kit with RT reagents. CleanPlex Indexed PCR Primers and CleanMag® Magnetic Beads are ordered separately to complete the workflow from input RNA to sequencing-ready NGS libraries. visit paragongenomics.com/store_mgi/

Product	SKU
CleanPlex for MGI SARS-CoV-2 Panel (8 reactions)	918001
CleanPlex for MGI SARS-CoV-2 Panel (96 reactions)	918002
CleanPlex for MGI SARS-CoV-2 Panel (384 reactions)	918003
CleanPlex for MGI Plated Single-Indexed PCR Primers, (96 indexes, 96 Rxns)	318012
CleanPlex for MGI Plated Single-Indexed PCR Primers, (96 indexes, 384 Rxns)	318013
CleanMag Magnetic Beads (5 mL)	718002
CleanMag Magnetic Beads (60 mL)	718003
MGISP-960 Automated Sample Preparation System	Variable

Learn More

To learn more about NGS applications for Infectious Diseases, visit www.paragongenomics.com/applications/infectious_disease/

To learn more about CleanPlex Technology, visit www.paragongenomics.com/cleanplex_technology/