

VirusHunter™ ARTIC SARS-CoV-2 Whole Genome Sequencing Assay

Highlights

- Competitive Pricing**
 Cost-effective, highly accurate sequencing of the full SARS-CoV-2 genome
- Fewer Steps and Hands-on Time**
 Capture-based sequencing technology eliminates tedious and time-consuming library preparation steps
- Proven Performance**
 Take advantage of new sequencing chips used in research labs across the United States and Asia
- High throughput**
 96-chip plate format enables the whole genome sequencing of 96 samples at once

Table 1: VirusHunter™ Sequencing Coverage and Accuracy

Ct Value	Coverage	Accuracy
<27	99.9%	99.98%
27-30	99%	99.9%
>30	98%	99%

Proven Technology with an Innovative Twist

Sequencing by hybridization exploits classic microarray technology with newer, smaller, more accurate chips. The VirusHunter™ ARTIC SARS-CoV-2 Whole Genome Sequencing Assay has been shown to outperform other sequencing technologies on the market¹.

The Need for Genomic Surveillance

As new mutations arise, SARS-CoV-2 whole genome sequencing is the only way to truly monitor the impact of the virus and emerging strains. GISAID and NCBI currently maintain enormous international public repositories of SARS-CoV-2 genomic sequences and epidemiological data analysis is available through NextStrain.

Knowledge is power. Sequencing is critical for epidemiological analyses and for making well-informed decisions for public health and safety. While most rare mutations are likely due to neutral evolution, emerging strains can be more transmissible and more lethal, and these strains could potentially evade vaccines².

Introduction

The VirusHunter™ ARTIC SARS-CoV-2 Whole Genome Sequencing Assay uses a multiplex tiling RT-PCR reaction originally developed by the ARTIC Network. These RT-PCR products are read out on QuadCore™ DNA sequencing chips with astounding accuracy. Core 0 of the QuadCore™ chip contains eight probes for each base in the SARS-CoV-2 genome; four probes (A, T, C, and G) on the sense strand, and four probes on the antisense strand. Cores 1-3 contain SARS-CoV-2 variant sequences and control sequences.

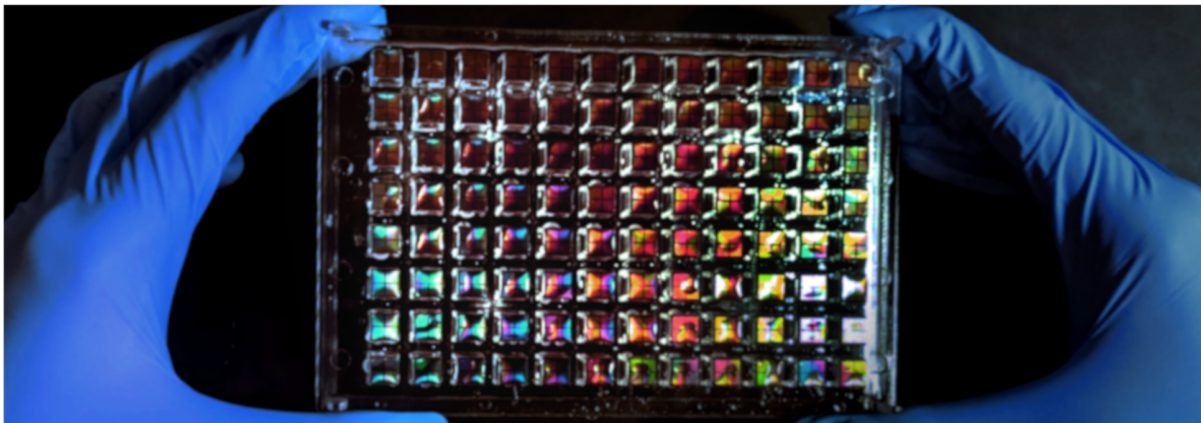


Figure 1: QuadCore™ DNA Sequencing chips arranged on a 96-chip plate

Workflow

Go from sample to sequence in under 24 hours with the VirusHunter™ ARTIC SARS-CoV-2 Whole Genome Sequencing Assay. Complete the sequencing workflow in six simple steps:

- RT-PCR (4.5 hours)
- Fragmentation (45 minutes)
- Hybridization (one hour or overnight)
- Staining (30 min)
- Scanning (8 minutes per chip)
- Data Processing (3 minutes per chip)

Simple, Intuitive Scanning

Setting up the Summit™ scanner takes under a minute per plate. The workflow is simple and intuitive. VirusHunter™ software outputs FASTA and FASTQ files for the SARS-CoV-2 whole genome, aligns the sequence to the reference, and creates a variant output file.

Variant Calling

Rapidly uncover important epidemiological and phylogenetic data with the VirusHunter™ variant calling output. The variant output file is automatically generated using VirusHunter™ software. It enables rapid sequence analysis and strain or clade determination using published variant data. Whole genome FASTA and FASTQ files are output at the same time to enable deeper sequencing analysis using custom pipelines.

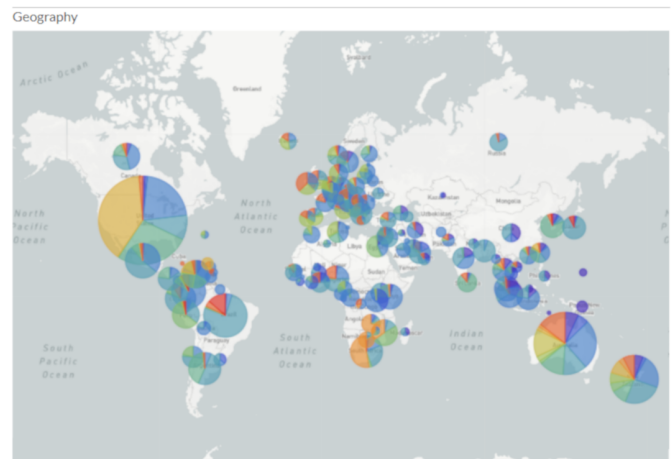
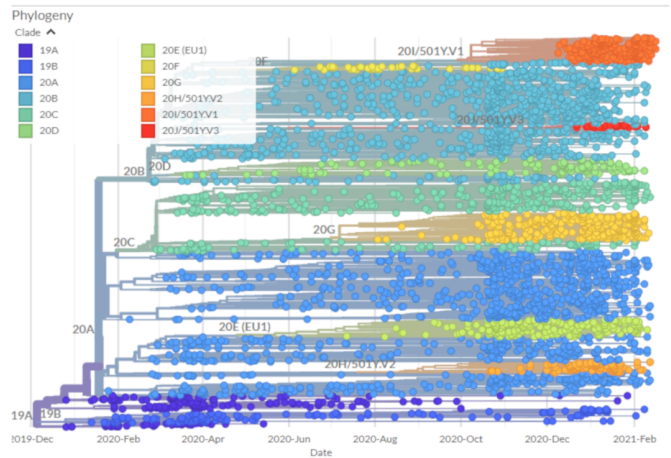


Figure 2: Genomic epidemiology and global subsampling of SARS-CoV-2 sequences from NextStrain[®].

Table 2: Variant Calling Output for B.1.1.7 (South African strain)

STRAIN	VARIANT MUTATION	VARIANT (NT)	START	END	REF SEQ	VARIANT SEQ	MEAN INTENSITY	MATCH	READ	HIT
B.1.1.7	aa:orf1ab:T1001I	C3267T	3267	3267	C	T	9.5985933	1	C	FALSE
B.1.1.7	aa:orf1ab:A1708D	C5388A	5388	5388	C	A	80.346773	1	C	FALSE
B.1.1.7	aa:orf1ab:I2230T	T6954C	6954	6954	T	C	14.283014	1	T	FALSE
B.1.1.7	del:11288:9	11288-11296 deletion	11288	11296	TCTGGTTTT		39.266993	9	TCTGGTTTT	FALSE
B.1.1.7	del:21765:6	21765-21770 deletion	21765	21770	TACATG		21.772854	6	TACATG	FALSE
B.1.1.7	del:21991:3	21991-21993 deletion	21991	21993	TTA		12.005803	3	TTA	FALSE
B.1.1.7	aa:S:N501Y	A23063T	23063	23063	A	T	7.6719067	1	A	FALSE
B.1.1.7	aa:S:A570D	C23271A	23271	23271	C	A	24.258374	1	C	FALSE
B.1.1.7	aa:S:P681H	C23604A	23604	23604	C	A	110.11712	1	C	FALSE
B.1.1.7	aa:S:T716I	C23709T	23709	23709	C	T	158.3074	1	C	FALSE
B.1.1.7	aa:S:S982A	T24506G	24506	24506	T	G	23.74348	1	T	FALSE
B.1.1.7	aa:S:D1118H	G24914C	24914	24914	G	C	16.618335	1	G	FALSE
B.1.1.7	aa:Orf8:Q27*	C27972T	27972	27972	C	T	123.8691	1	C	FALSE
B.1.1.7	aa:Orf8:R52I	G28048T	28048	28048	G	T	38.490501	1	G	FALSE
B.1.1.7	aa:Orf8:Y73C	A28111G	28111	28111	A	G	58.888179	1	A	FALSE
B.1.1.7	aa:N:D3L	28280 GAT->CTA	28280	28282	GAT	CTA	18.418404	3	GAT	FALSE
B.1.1.7	aa:N:S235F	C28977T	28977	28977	C	T	27.874411	1	C	FALSE

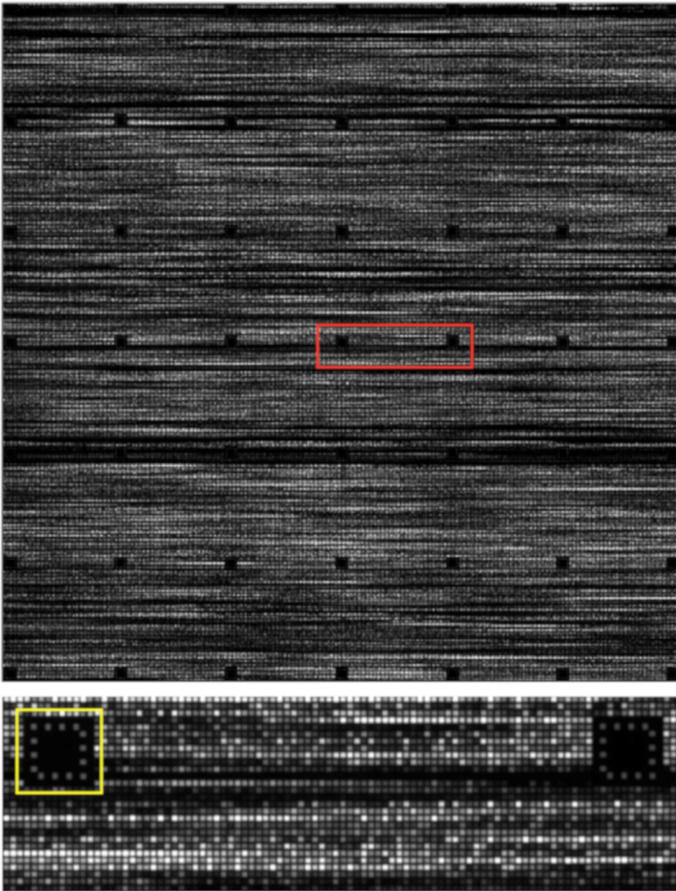


Figure 3: Core of a QuadCore™ DNA Sequencing chip after hybridization. The area highlighted in red has been enlarged in the second image. The area highlighted in yellow is an alignment mark, a control sequence repeated across the chip and used for image processing and analysis.

References

1. Hoff, Kendall, et al. "Highly Accurate Chip-Based Resequencing of SARS-CoV-2 Clinical Samples." *Langmuir* (2021).
2. Thomson, E. C. et al. Circulating SARS-CoV-2 spike N439K variants maintain fitness while evading antibody-mediated immunity. *Cell* (2021) doi:10.1016/j.cell.2021.01.037.
3. Nextstrain / nCoV / global.
<https://nextstrain.org/ncov/global>.



Figure 4: The Summit™ Scanner

Summary

The VirusHunter™ ARTIC SARS-CoV-2 Whole Genome Sequencing Assay breaks sequencing barriers by enabling, faster, more accurate, and less-expensive whole genome sequencing, which in turn enables visionary scientists to study, monitor, and change outcomes during a world-wide pandemic. The VirusHunter™ system provides the precision to answer the most important questions scientists are asking today about the SARS-CoV-2 virus.

Learn More

To learn more about the VirusHunter™ ARTIC SARS-CoV-2 Whole Genome Sequencing Assay, visit www.centrilliontech.com/virushunter.