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DATA SHEET

Rapid Viral Surveillance Panel v2

Streamlined whole-genome sequencing for high-risk viral surveillance and research

Expanded panel provides coverage of ~200 viruses, including those of public health concern[1–6]

Hybrid–capture enrichment accommodates RNA and DNA viral pathogens

Rapid workflow supports a range of host and environmental sample types[7]

Identifying high-impact viruses for public health surveillance

Viral genome surveillance plays a pivotal role in global health security by providing invaluable insights into the evolution, spread, and activity of pathogens.[1] Analyzing the genetic makeup of viruses using next-generation sequencing (NGS) allows scientists to track mutations that may affect transmissibility, virulence, or resistance to treatment.[8] This information is crucial for designing effective diagnostic tests, therapeutics, and vaccines to combat emerging infectious diseases.

The Illumina Rapid Viral Surveillance Panel v2 is an NGS panel that enables the detection and whole-genome sequencing (WGS) of ~200 viruses, including viruses identified as important risks to public health (Table 1).[1–6] The panel uses a hybrid–capture target enrichment workflow that allows for sequencing of various sample types in as little as 24 hours without the high read depth required by shotgun metagenomics methods. Compared to other targeted resequencing methods, such as amplicon sequencing, hybrid–capture provides more uniform coverage across viral genomes and a greater ability to identify mutations and divergent sequences, making the Viral Surveillance Panel v2 ideal for outbreak surveillance and variant monitoring.

Streamlined NGS workflow

The Rapid Viral Surveillance Panel v2 workflow enriches for viral genomes from a range of sample types, including wastewater, serum, plasma, skin lesions, and nasopharyngeal swabs.[7] Libraries are prepared from RNA or DNA extracted from host or environmental samples, sequenced on an Illumina benchtop sequencing system, and analyzed using the DRAGEN™ Microbial Enrichment Plus App available on BaseSpace™ Sequence Hub. The library preparation and sequencing steps can be completed in as little as 24 hours with minimal hands-on time (Figure 1).[7]

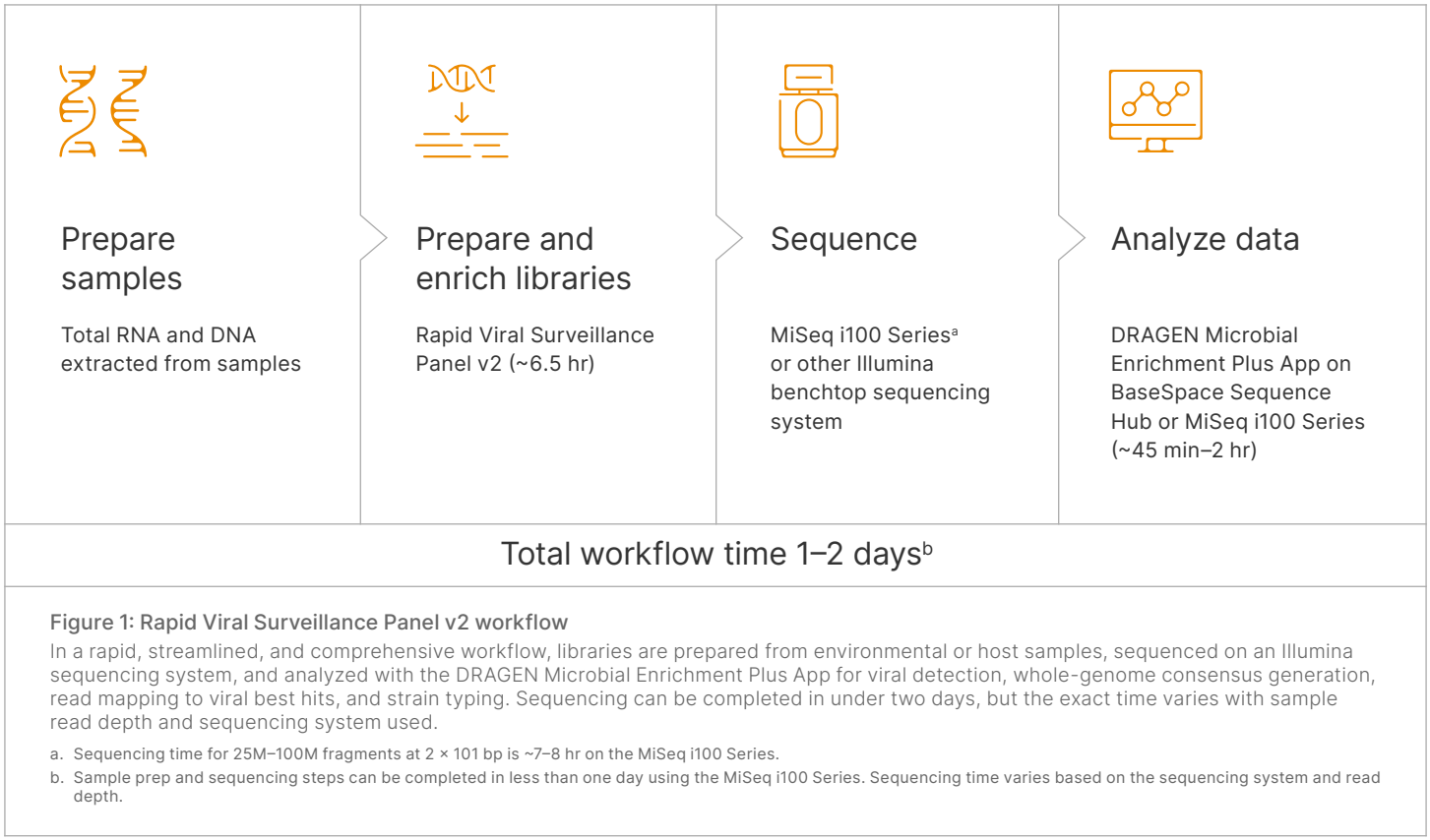

Figure 1: Rapid Viral Surveillance Panel v2 workflow
In a rapid, streamlined, and comprehensive workflow, libraries are prepared from environmental or host samples, sequenced on an Illumina sequencing system, and analyzed with the DRAGEN Microbial Enrichment Plus App for viral detection, whole-genome consensus generation, read mapping to viral best hits, and strain typing. Sequencing can be completed in under two days, but the exact time varies with sample read depth and sequencing system used.

a. Sequencing time for 25M–100M fragments at 2 × 101 bp is ~7–8 hr on the MiSeq i100 Series.
b. Sample prep and sequencing steps can be completed in less than one day using the MiSeq i100 Series. Sequencing time varies based on the sequencing system and read depth.

Table 1: Key high-risk viruses included on the Rapid Viral Surveillance Panel v2

Adeno-associated virus 2	Human adenovirus A–G	Mayaro virus	Sabia virus
Aichi virus 1	Human bocavirus	Measles virus	Salivirus A
Aigai virus	Human coronavirus	Menangle virus	Sandfly fever Sicilian virus
Bombali virus	Human cytomegalovirus	Middle East respiratory syndrome–related coronavirus	Sapovirus
Bourbon virus	Human immunodeficiency virus 1/2	Mpox virus	Severe acute respiratory syndrome-related coronavirus
Cache Valley virus	Human metapneumovirus	Mumps virus	Severe acute respiratory syndrome-related coronavirus 2
California encephalitis virus	Human papillomavirus	Murray Valley encephalitis virus	Semliki Forest virus
Chapare virus	Human parainfluenza virus 1–4	Nipah virus	Severe fever with thrombocytopenia syndrome virus
Chikungunya virus	Human parechovirus	Norovirus	Sindbis virus
Colorado tick fever virus	Human parvovirus B19	Omsk hemorrhagic fever virus	Snowshoe hare virus
Coxsackievirus A/B	Influenza virus A–C	O'nyong-nyong virus	Sosuga virus
Crimean-Congo hemorrhagic fever virus	Jamestown Canyon virus	Oropouche virus	St. Louis encephalitis virus
Dengue virus 1–4	Japanese encephalitis virus	Poliovirus	Tacheng tick virus 2
Ebola virus	Junin virus	Polyomavirus	Tahyna virus
Echovirus	Kyasanur Forest disease virus	Powassan virus	Tick-borne encephalitis virus
Enterovirus A–D	La Crosse virus	Punta Toro virus	Torque teno virus
Epstein-Barr virus	Lassa virus	Rabies virus	Toscana virus
Equine encephalitis virus	Lloviu virus	Ravn virus	Usutu virus
Guanarito virus	Lujo virus	Respiratory syncytial virus A/B	Varicella-zoster virus
Hantavirus	Lymphocytic choriomeningitis virus	Rhinovirus A–C	Variola virus
Heartland virus	Lyssavirus	Rift Valley fever virus	West Nile virus
Henipavirus	Machupo virus	Ross River virus	Yellow fever virus
Hepatovirus A–E	Mamastrovirus	Rotavirus A/B/C/H	Zika virus
Herpes simplex virus 1/2	Marburg virus	Rubella virus	

Library preparation

The Rapid Viral Surveillance Panel v2 library preparation workflow consists of pre-enrichment and enrichment steps. The rapid pre-enrichment library step is greatly simplified and streamlined to be performed in half the time, with a reduced number of steps, compared to the nonrapid format. Pre-enrichment generates hundreds of thousands of nontargeted libraries that are enriched with Viral Surveillance Panel v2 probes using a hybrid–capture approach. Enrichment with on-bead tagmentation provides a rapid, automation-compatible workflow that can be completed with minimal hands-on time. The protocol accommodates sample input amounts ranging from 10 ng to 100 ng total nucleic acid and supports multiplexing of up to 384 samples in a single run.

Sequencing

The lower read depth requirements for libraries enriched with Viral Surveillance Panel v2 allow for multiple sequencing system options, including the benchtop MiniSeq™, MiSeq™ i100 Series, NextSeq™ 550, NextSeq 1000, and NextSeq 2000 Systems. Viral titer, nucleic acid sample quality, sample read depth, and the number of reads per sample impact the number of virus-specific reads and sequence coverage obtained. The general sequencing read depth recommendation for good quality samples is a minimum of 1M fragments per sample with a read length of 2 × 150 bp. The recommended sample read depth also varies with sample type. For more complex samples, such as wastewater, a minimum of 4M fragments per sample are recommended. Abundant off-target reads are expected if other microbial nucleic acids are present, such as from bacteria found in complex sample types.

Data analysis

Data generated using the Viral Surveillance Panel v2 are analyzed using the DRAGEN Microbial Enrichment Plus App available on BaseSpace Sequence Hub and onboard MiSeq i100 Series Systems. This easy-to-use analysis pipeline provides sample quality control, reference-guided alignment to a broad, curated viral genome database, variant calling, viral genome consensus sequence generation, antiviral resistance prediction for Influenza A/B viruses, flexible reporting options, and integration with Pangolin and Nextclade for further phylogenetic assignment of supported viruses.

Performance

Target enrichment

Compared to shotgun metagenomic sequencing, where all RNA or DNA is sequenced, targeted hybrid–capture using the Viral Surveillance Panel v2 minimizes unnecessary sequencing of host and nontargeted microbes, reducing costs and allowing for broad sequencing of viral genomes on benchtop sequencing systems.[7]

Viral genome recovery using enrichment with the Viral Surveillance Panel v2 was compared with shotgun metagenomic sequencing without enrichment. Analysis was performed using wastewater samples collected through collaborations with Colorado State University (n = 29) and Wisconsin State Lab of Hygiene (n = 32). The Viral Surveillance Panel v2 demonstrated superior viral genome recovery from multitarget contrived samples compared to shotgun metagenomic sequencing (Figure 2).

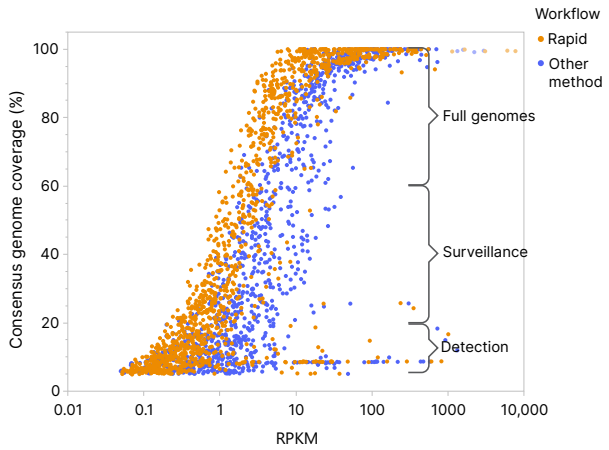

Figure 2: Rapid Viral Surveillance Panel v2 performance compared to shotgun metagenomic sequencing

Rapid Viral Surveillance Panel v2 (Rapid) produces higher genome coverage measured as reads per kilobase of transcript per million reads mapped (RPKM) compared to shotgun metagenomic RNA sequencing with another commercially available method. Increased coverage of targeted genomes Rapid Viral Surveillance Panel v2 ensures more full genomes are captured and delivers excellent performance for detection and surveillance. Analysis was performed using wastewater samples collected through collaborations with Colorado State University (n = 29) and Wisconsin State Lab of Hygiene (n = 32).

Wastewater surveillance

Surveillance for viral sequences in wastewater provides a regional indicator of communal spread of viral pathogens, giving public health professionals valuable information for response planning.[8] The Viral Surveillance Panel can be used with these samples to enable early detection and identification of viral genomes in wastewater at lower concentrations than shotgun sequencing (Table 2).

Wastewater samples from two collection sites extracted through collaborations with the Wisconsin State Lab of Hygiene (WSLH) and Colorado State University (CSU). Three samples from each collection site were assessed. Libraries prepared from these six wastewater samples were sequenced and normalized to 4M fragments for Viral Surveillance Panel v2 enrichment or 4M and 25M fragments for shotgun metagenomic sequencing. A sequencing depth of 4M fragments was used in this comparison because wastewater samples can vary greatly in complexity and may contain dozens of viruses at low abundance. The Viral Surveillance Panel v2 demonstrated increased viral detection sensitivity in a complex environmental sample type with low overall viral load compared to shotgun metagenomic sequencing, even when total reads for shotgun metagenomic sequencing were increased ~six-fold (Table 2).

Table 2: Top viruses detected in wastewater using Rapid Viral Surveillance Panel v2 or shotgun metagenomic sequencing

Virus (strain)	4M fragments				25M fragments	
	Rapid Viral Surveillance Panel v2		Shotgun metagenomics		Shotgun metagenomics	
	Genome coverage (%)	Read count	Genome coverage (%)	Read count	Genome coverage (%)	Read count
Sapovirus (GII.1)	99.7	219,539	50.0	57	84.2	360
Human adenovirus F (Human adenovirus 41)	100	104,693	6.4	18	23.6	72
Human coronavirus OC43 (HCoV_OC43)	98.1	23,857	0	0	10.0	26
Sapovirus (GV)	99.6	10,750	0	0	25.3	19
Human adenovirus E	88.4	6733	0	0	0	0
JC polyomavirus (JCPyV)	99.3	5834	0	0	0	0
Mamastrovirus 9 (MAstV9)	99.2	4959	0	0	0	0
Mamastrovirus 1 (MAstV1)	98.6	3972	7.5	5	22.0	13
Human adenovirus A (Human adenovirus 31)	81.1	3449	0	0	0	0
Mamastrovirus 6 (MAstV6) [MLB1]	97.2	3181	0	0	17.3	9
Norovirus (G1)	96.9	1972	0	0	0	0
BK polyomavirus (BKPyV)	100	1522	0	0	11.1	4
Mamastrovirus 8 (MAstV8) [VA2]	92.1	1208	0	0	6.1	4
Human papillomavirus 59 (HPV59; high-risk)	69.3	1015	0	0	0	0
Enterovirus A (not Coxsackievirus) [Enterovirus A71]	70.0	295	5.1	4	9.0	6

Summary

The Rapid Viral Surveillance Panel v2 is part of an optimized, comprehensive workflow for detecting viral outbreaks, zoonotic surveillance, and tracking mutations. This kit uses updated chemistry to deliver the same high-quality results as the existing Viral Surveillance Panel v2 with a faster turnaround time as little as 24 hours for time sensitive applications.[7] The streamlined workflow is compatible with a range of sample types and applications, including biological research samples and wastewater surveillance for regional presence of viruses.

The kit includes hybrid–capture probes for identifying ~200 RNA and DNA virus genomes that have been designated as high risks to public health. The hybrid–capture target enrichment minimizes the need for high sample read depth by focusing on target sequences, reducing costs while increasing throughput. Data generated using the Rapid Viral Surveillance Panel v2 can be analyzed with the user-friendly DRAGEN Microbial Enrichment Plus App onboard the MiSeq i100 Series or online through BaseSpace Sequence Hub. This robust NGS workflow delivers excellent viral capture performance for identifying DNA and RNA in complex samples, providing public health organizations and researchers with an advanced alternative to shotgun sequencing.

Learn more →

Rapid Viral Surveillance Panel v2 (link pending)

DRAGEN Microbial Enrichment Analysis Plus App

Illumina sequencing systems

Ordering information

Product	Catalog no.
Illumina Rapid Viral Surveillance Panel v2 Kit (96 samples)	20158826

References

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