Aplex BIO

PCR hyperplexed without compromise

Democratizing high-performance multipathogen surveillance with next-gen Hyperplex PCR™

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Chief Scientific Officer

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Company and Purpose

- Founded in 2020
- Headquartered in Karolinska Institutet Campus, Stockholm
- Formed by industry and academic leaders in Molecular Diagnostics behind companies like Olink, Cartana, ParAllele Bioscience, Qlinea, and Halo Genomics

Aplex BIO

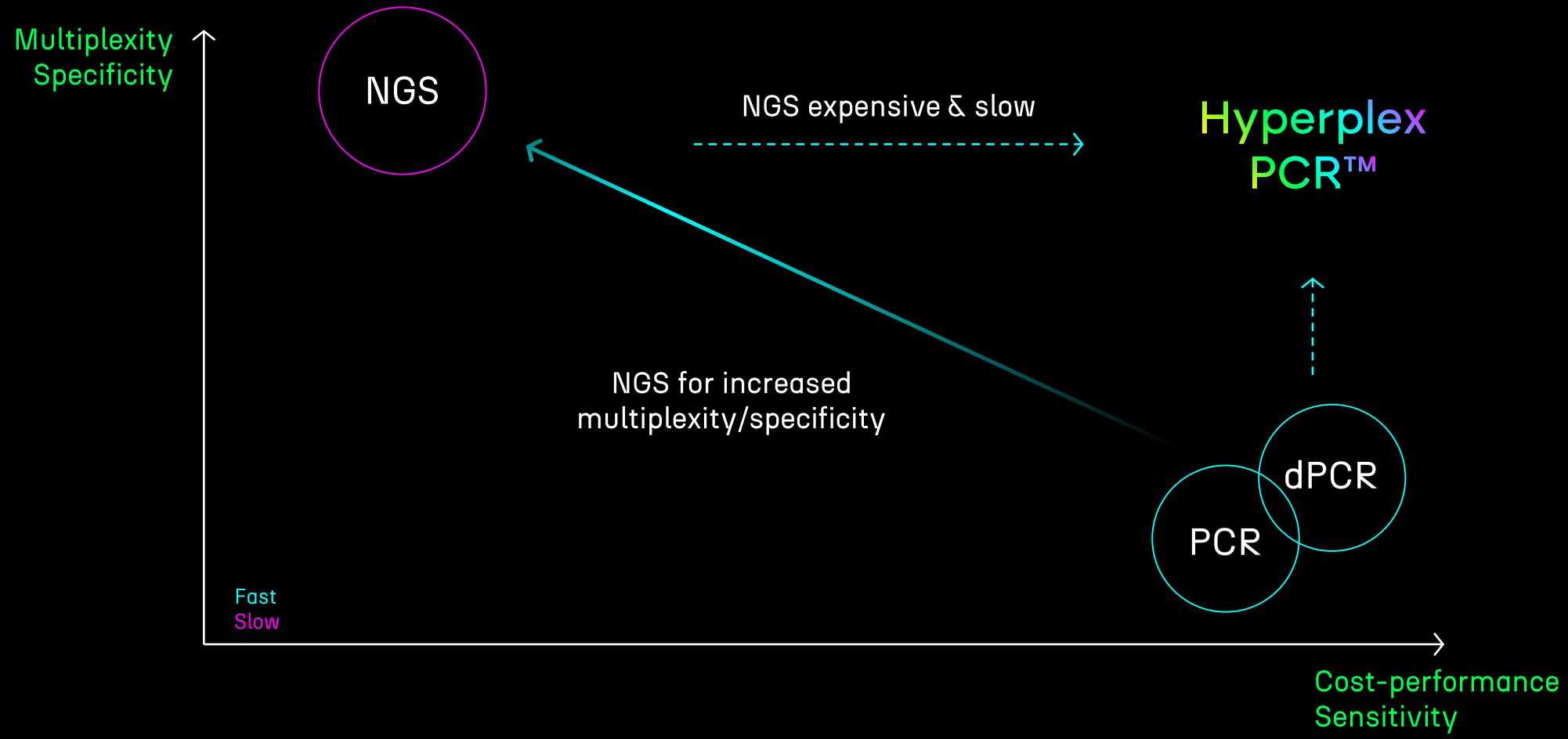
Mission

To push the boundaries of molecular analysis enabling unprecedented multiplexing, sensitivity and specificity without compromise

Vision

To be a globally recognized leader in molecular analysis technology, providing multiple, gold-standard hyperplexing solutions for healthcare and sustainability

Unmet Need In Efficient Molecular Diagnostics



Introducing Hyperplex PCR™

Multiplexity of 100+

No splitting, no sequencing

New probes on-demand

With NGS-grade specificity, no optimization

High & low abundance targets

Simultaneous detection in one tube

Reproducible & Precise

With state-of-the-art CV in every assay

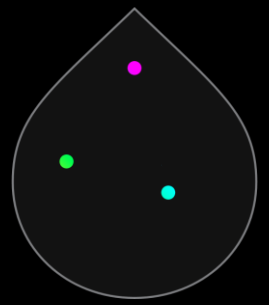
Affordable & fast

With results within 1 day

Single-molecule sensitivity

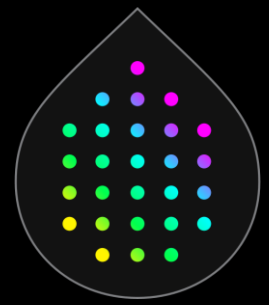
Ability to count PCR amplicons

Hyperplex PCR™ - Multiplexing Unleashed



Conventional PCR
3-5 biomarkers

Limited information
split sample or rely on sequencing



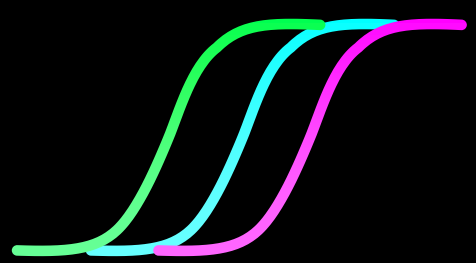
Hyperplex PCR
100+ biomarkers

Complete picture
unparalleled simple solution

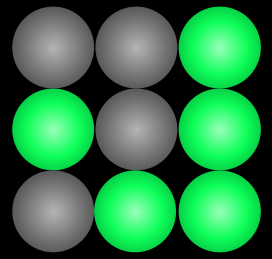
With Cutting-Edge Nanoprobes and Molecular Biology

Based on Over 20 Years of Research

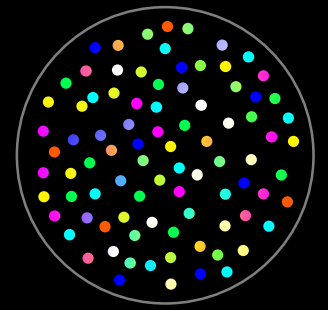
We Have Taken PCR to a Whole New Level



qPCR



Digital PCR

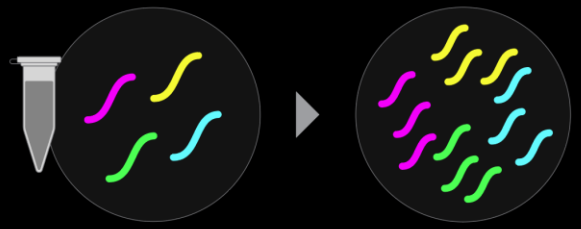


Hyperplex PCR™

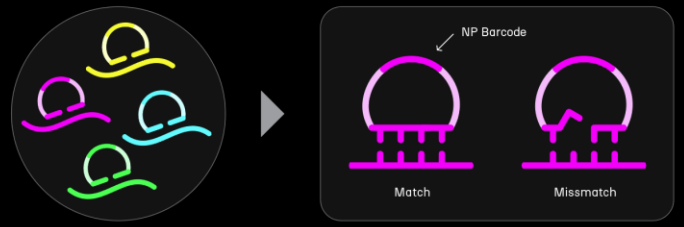
Multiplex/well	≤5	6	100+
LoD (copies/reaction)	~5	1	1
Single-nucleotide specificity	X	-	✓
Simple probe design	X	X	✓
Cost	$	$$	$
Counts	-	10,000	1,000,000

Hyperplex PCR™ Assay

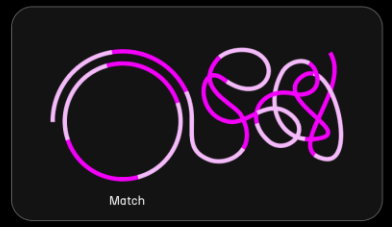
01 PCR Amplification



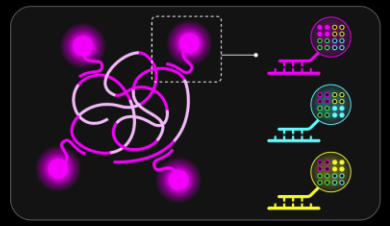
02 Padlock probing & Ligation



03 Rolling-Circle Amplification

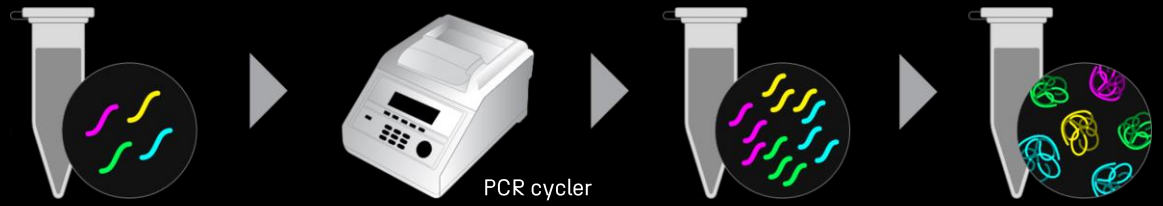


04 Nanopixel Labelling



Simple Workflow, Common Instruments

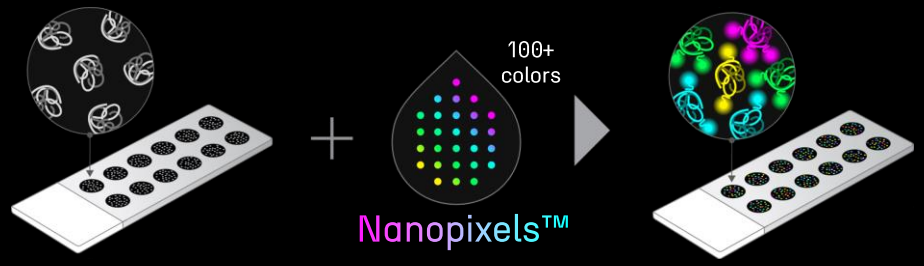
hpPCR assay - PCR/Ligation/RCA



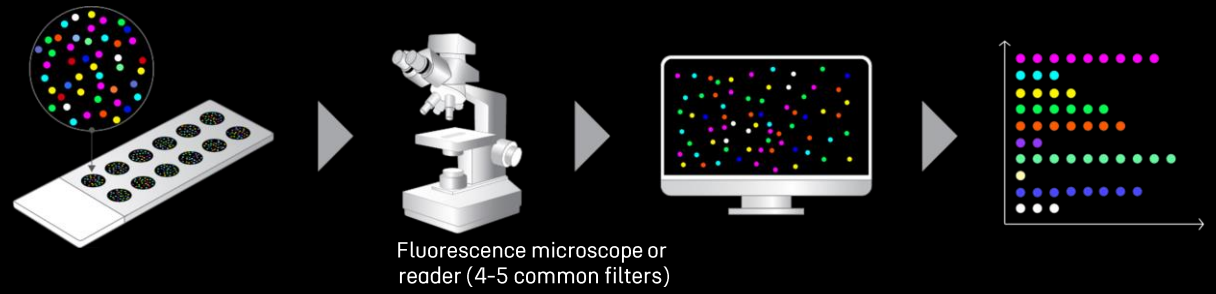
Mixed sample
>100 markers

Compatible with any
off-the-shelf extraction kit

Sample capture - slide or plate



Fluorescence imaging and automated image analysis



>100-plex output
in a single step

Turnover time
< 1 day

hpPCR used for COVID WW monitoring in Sweden

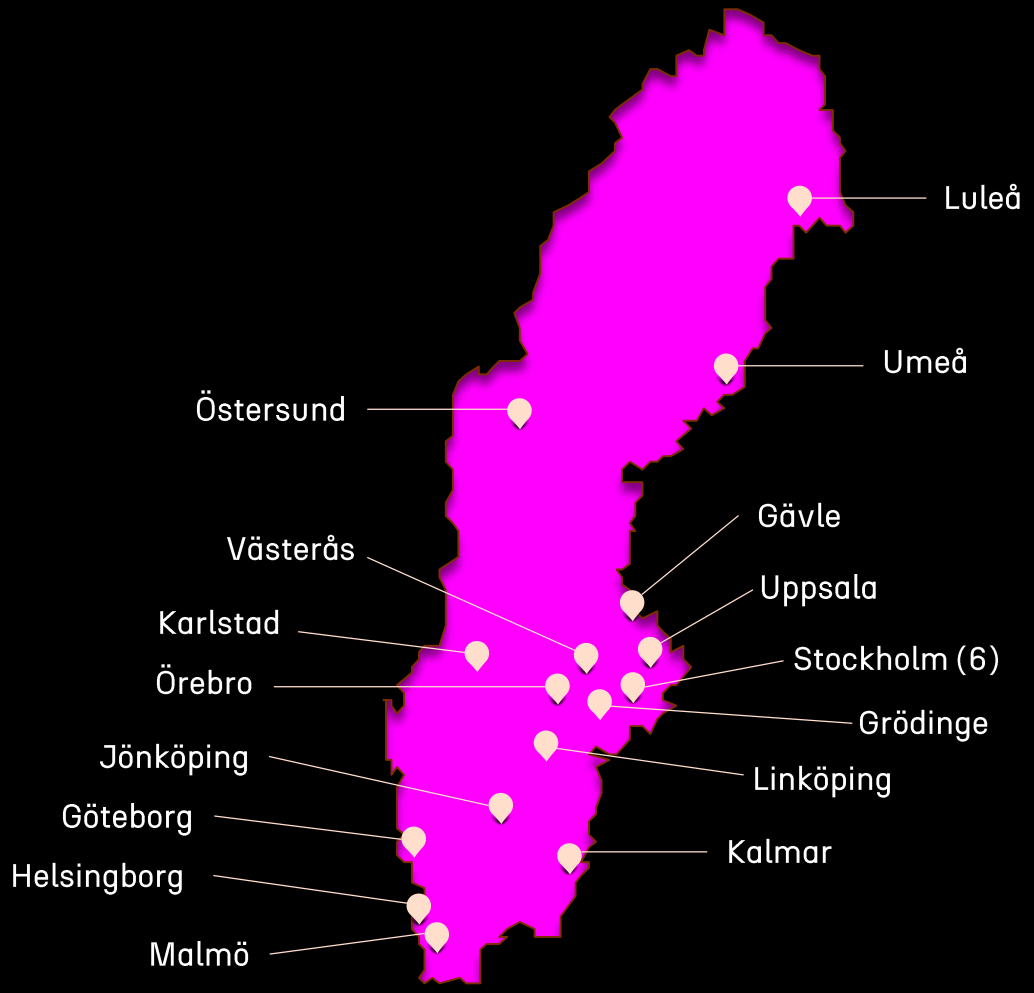
on behalf of Swedish Environmental Epidemiology Center (SEEC)

Powered by Nanopixels™

18

Targets Monitored

N3	S:T547I
S:R346T	S:Q613H
S:K444T	Orf9b:I5T
S:N460K	Orf9b:N55S
S:F490S	S:483del
S:F456L	S:F157S/R158G
S:Q52H	orf1:A7842G
S:F486P	S:69-70del
S:T478R	PMMoV



22

Sites Monitored

On weekly basis.

Results are delivered within 3 days.

Application note released on our website

https://www.aplex.bio/application-note-wbe

Hyperplexed Variant Surveillance

COVID Monitoring
Oct '22-Nov '23

700+

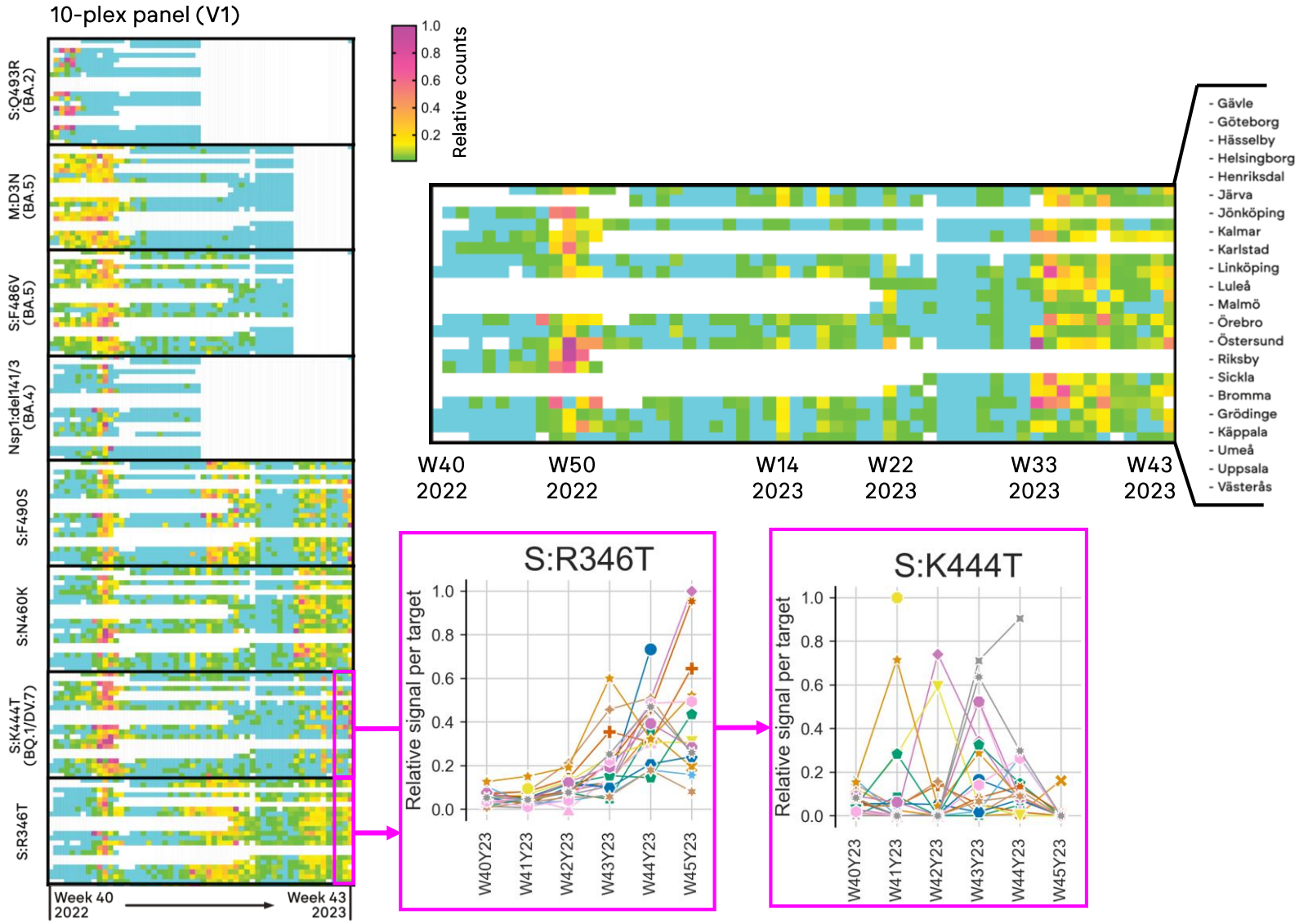
Samples analysed (duplicates)

10,000+

Data points

18-plex+

Per sample, modified on demand



Hyperplexed Variant Surveillance

COVID Monitoring Oct '22-Nov '23

700+

Samples analysed (duplicates)

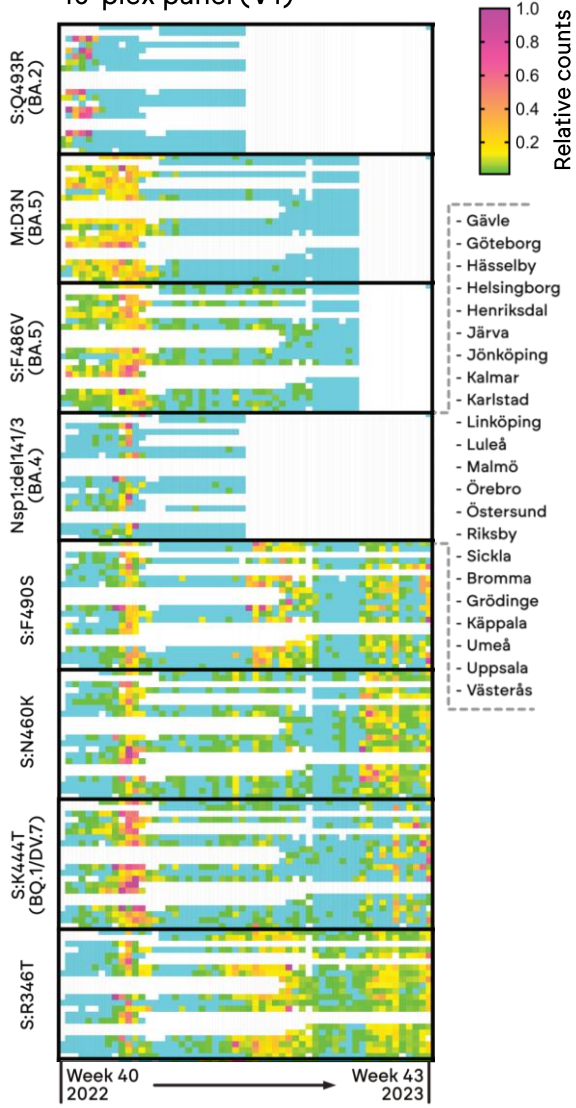
10,000+

Data points

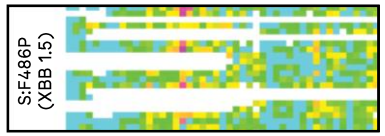
18-plex+

Per sample, modified on demand

10-plex panel (V1)

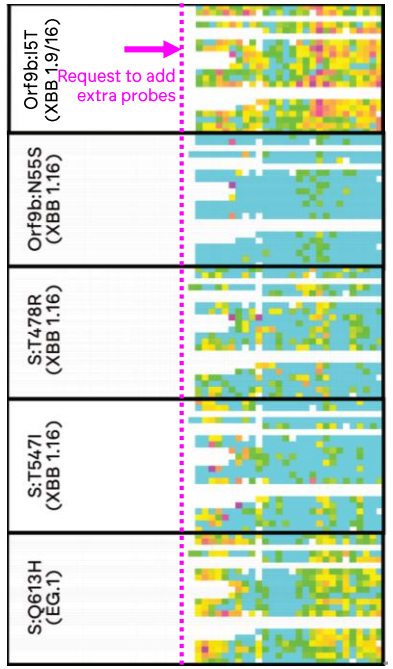


11-plex panel (V2)



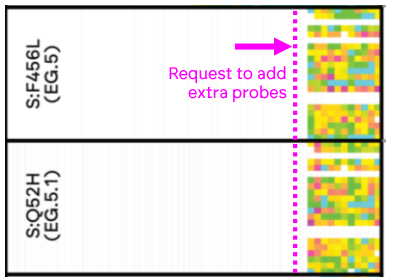
Additional probe for S:F486P (XBB 1.5) requested W49/2022

14-plex panel (V3)



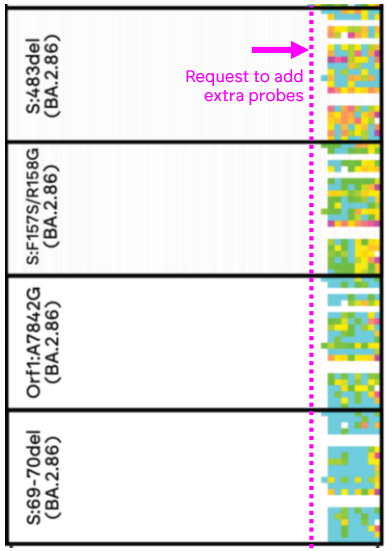
Additional probes for mutations characteristic of XBB 1.9/16 and EG.1
BA.2+BA.4 probes excluded

16-plex panel (V4)



Probes for BA.5 excluded

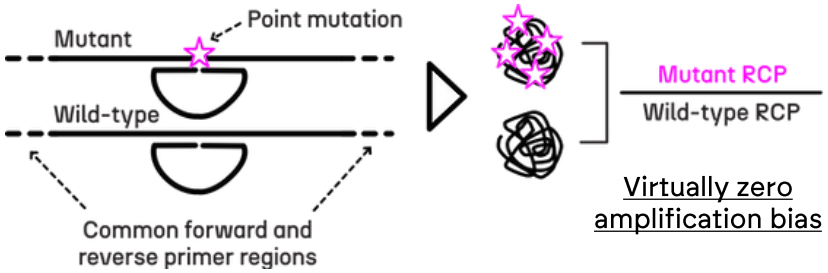
18-plex panel (V5)



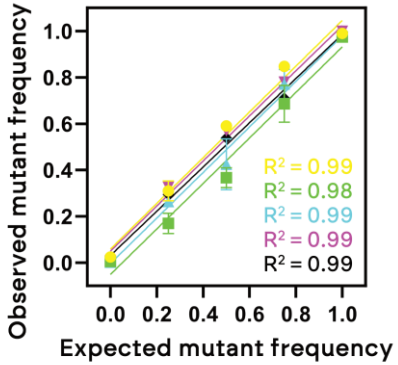
All panel modifications carried out within 2-weeks

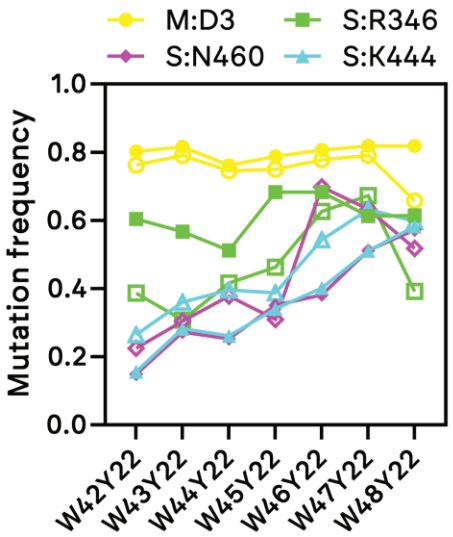
Mutation Frequency and Earlier Detection

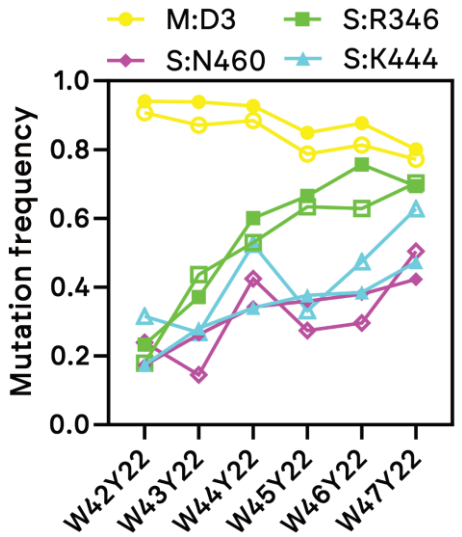
hpPCR allows precise and sensitive quantification of mutation frequency in wastewater samples

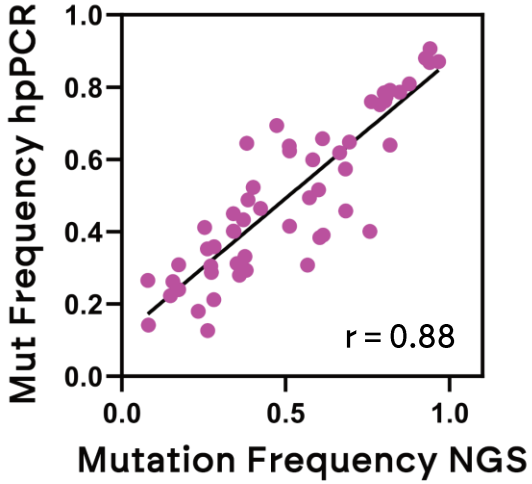


Virtually zero amplification bias

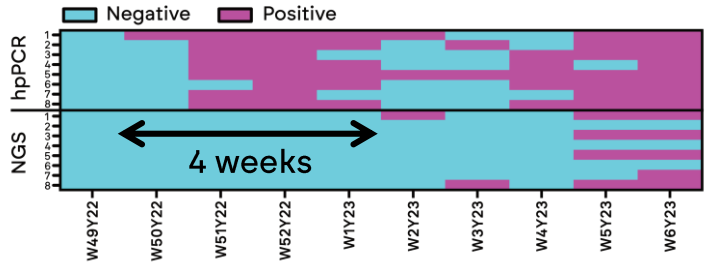








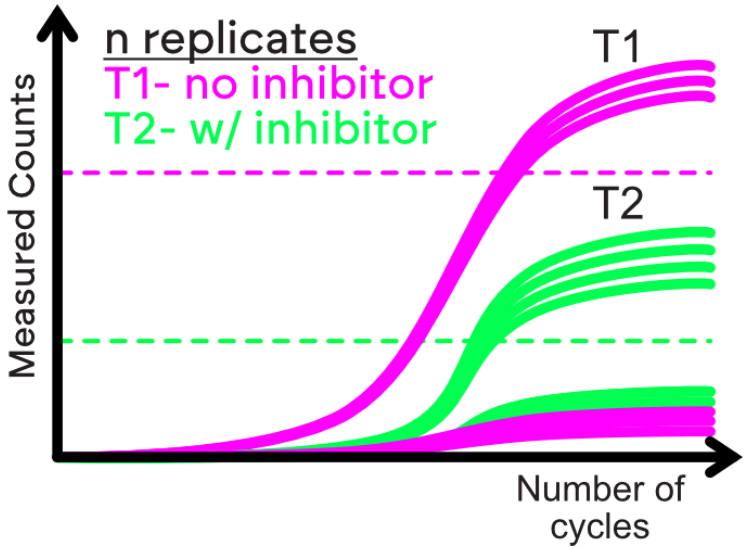
hpPCR allows early detection of emerging variants vs NGS at least 4 weeks earlier



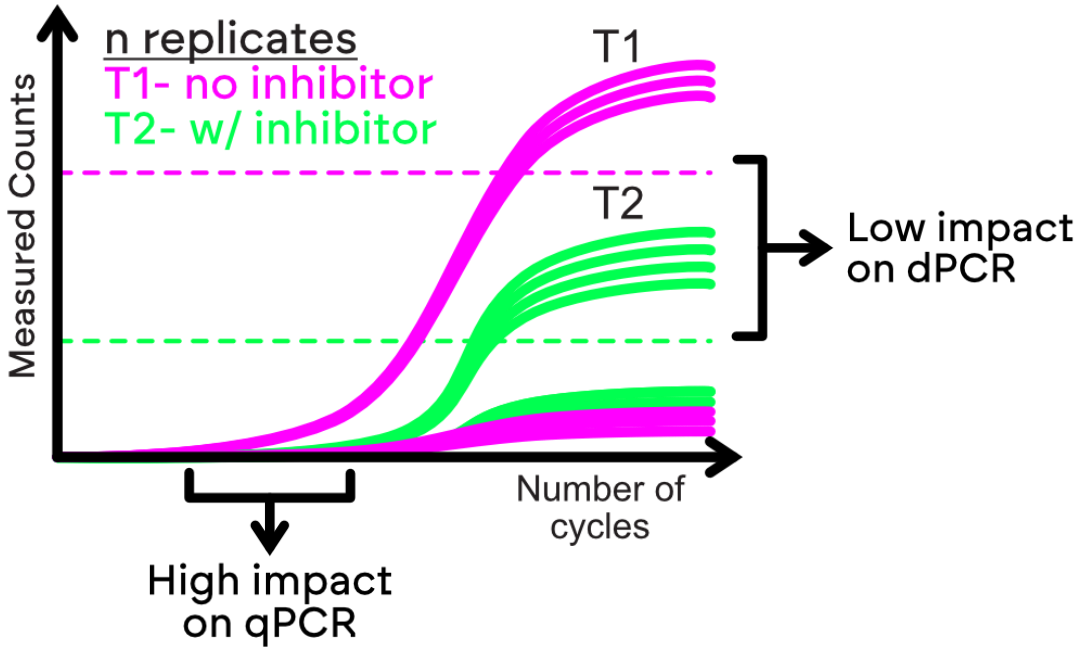
Weekly measurement of S:F486P, characteristic of XBB 1.5 using hpPCR and NGS (Ion torrent). 8 wastewater collection sites were measured at each time point.

Both measurements were performed on the same extracted nucleic acid sample.

High Resistance to Inhibition

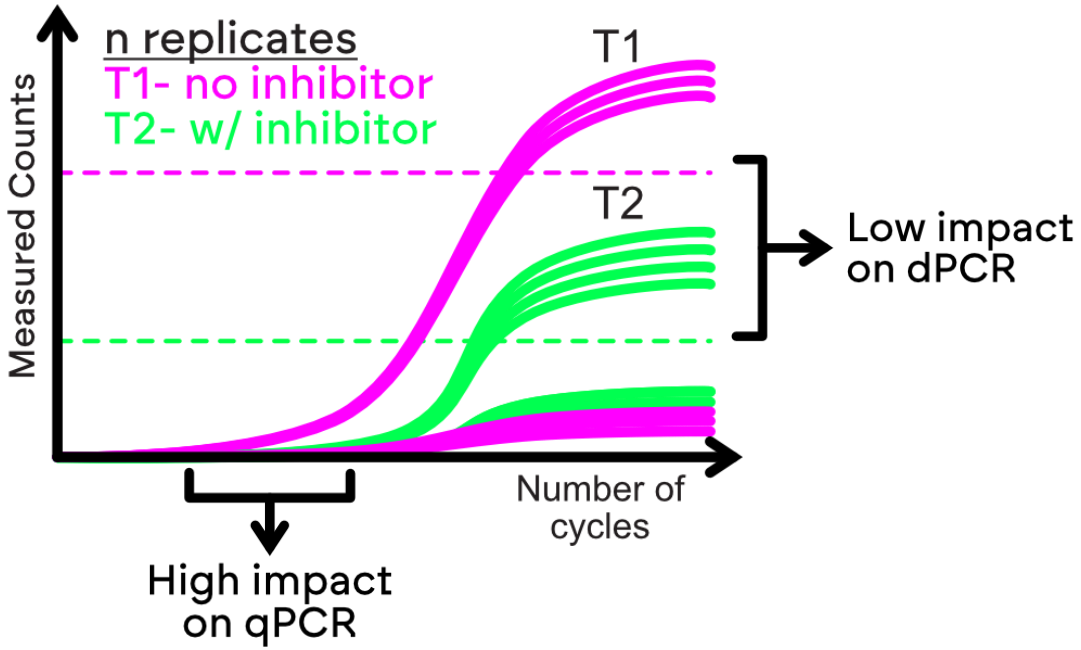


High Resistance to Inhibition

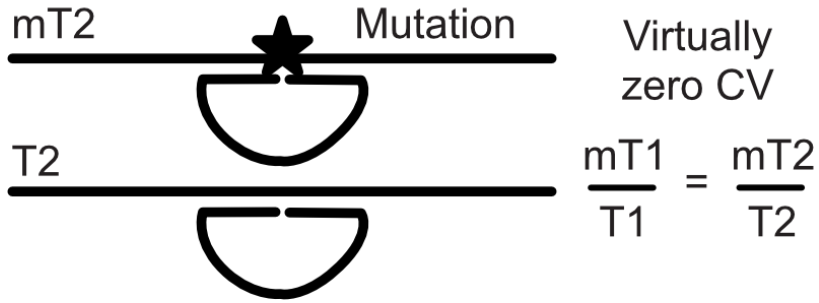


High Resistance to Inhibition

Mutation frequency readout and
internal controls neutralize PCR bias

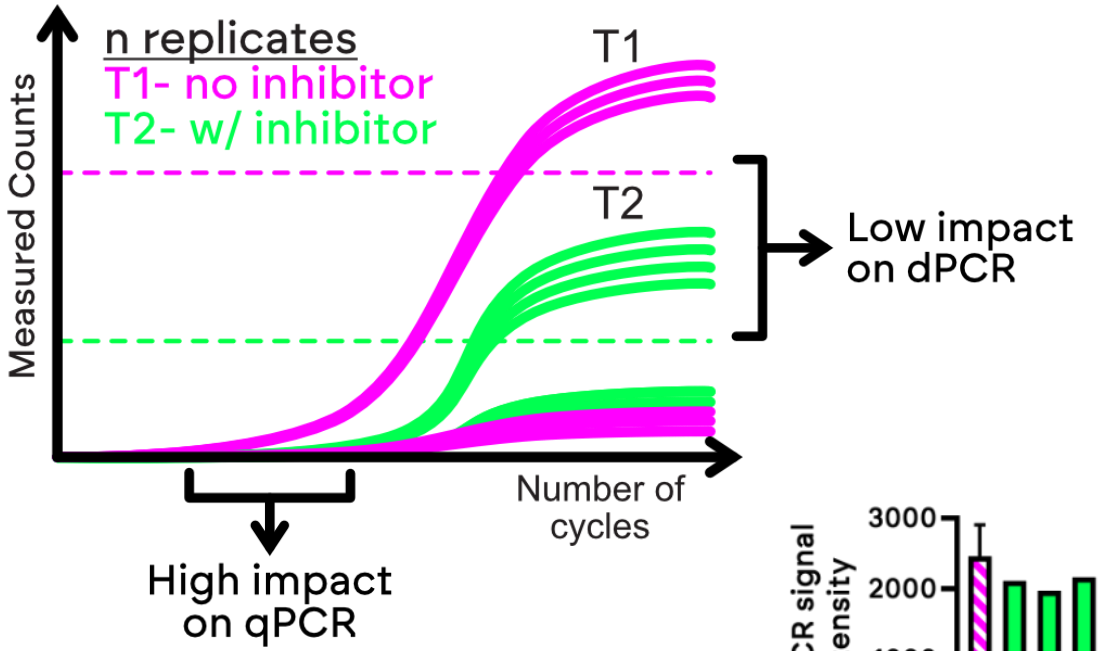


Virtually zero impact on Hyperplex PCR

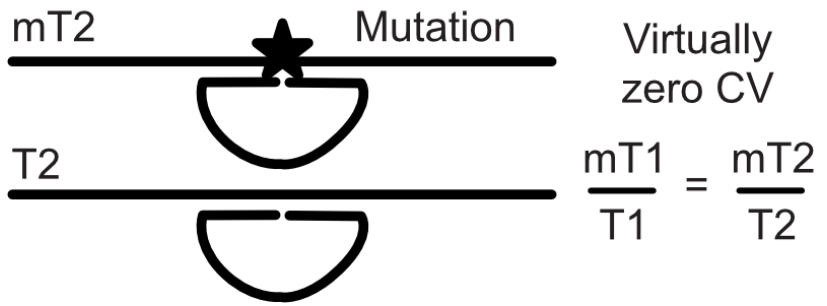


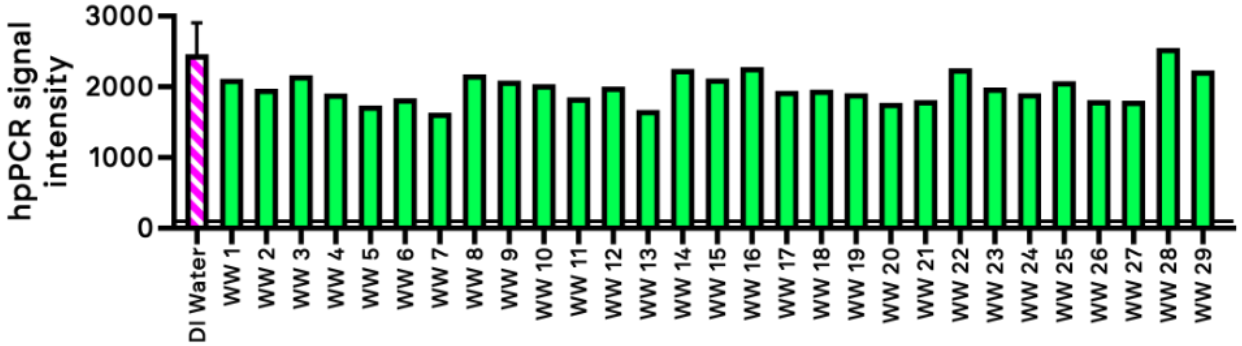
High Resistance to Inhibition

Mutation frequency readout and internal controls neutralize PCR bias + Intensity of RCA signals independent of inhibition in wastewater samples



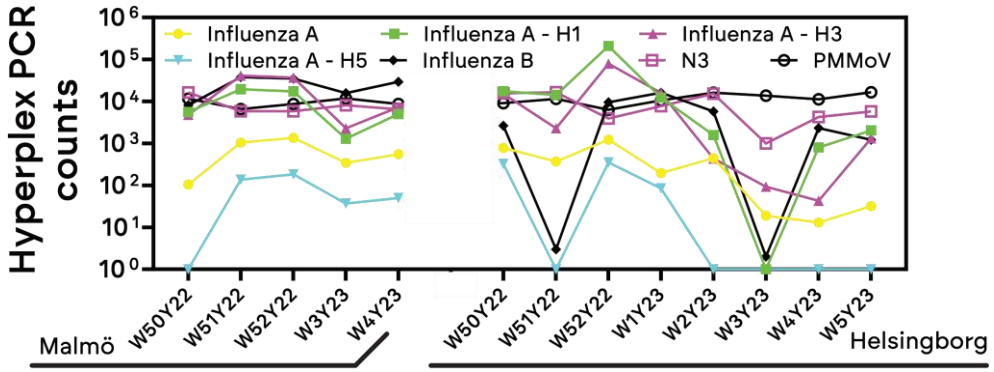
Virtually zero impact on Hyperplex PCR





Quantitative Multi-Pathogen Analysis

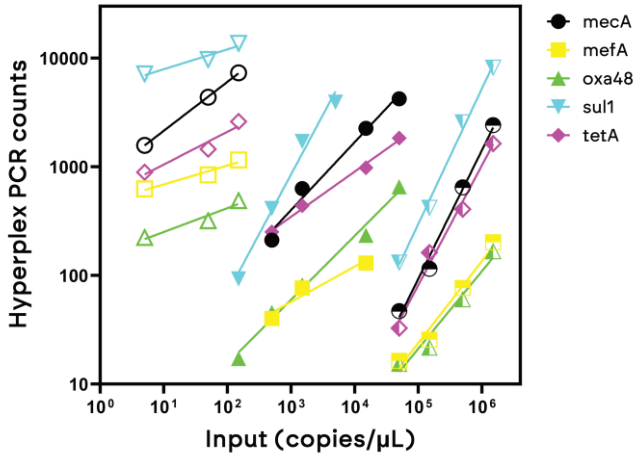
- Versatile multi-pathogen panels
  - Influenza
  - AMR
  - SARS-CoV-2



Monitoring of wastewater in two sites using a 7-plex multi-pathogen / multi-subtype panel.

All plotted signals are significantly above the blank baseline by at least 3 standard deviations. 10^0 means non-detection.

- Absolute quantification
- Dynamic range >6 orders
- High & low abundance in same sample



Site	mecA (cop/µL)	mefA (cop/µL)	oxa48 (cop/µL)	sul1 (cop/µL)	tetA (cop/µL)
1	< 10	$1.1x10^4$	< 10	$8.9x10^4$	$1,5x10^3$
2	< 10	$1.2x10^4$	< 10	$1.3x10^5$	$1,7x10^3$
3	< 10	$1.2x10^4$	< 10	$1.2x10^5$	$5,3x10^3$
4	$1.2x10^2$	$2.0x10^4$	< 10	$4.7x10^5$	$4,9x10^3$
5	48	$3.4x10^5$	< 10	$2.8x10^5$	$1,4x10^4$
6	$2,2x10^2$	$7.1x10^4$	< 10	$6.7x10^4$	$4,2x10^3$
7	$1,2x10^3$	$1.0x10^4$	< 10	$2.3x10^5$	$3,3x10^4$
8	$9,8x10^2$	$9.8x10^3$	< 10	$8.3x10^4$	$2,6x10^4$
9	$5.8x10^3$	$9.8x10^2$	< 10	$2.4x10^5$	$7,8x10^3$
10	< 10	$5.0x10^2$	N.D.	$2.6x10^5$	$6,1x10^2$

Summary

Hyperplex PCR™ – The Ultimate Tool for Pandemic Preparedness

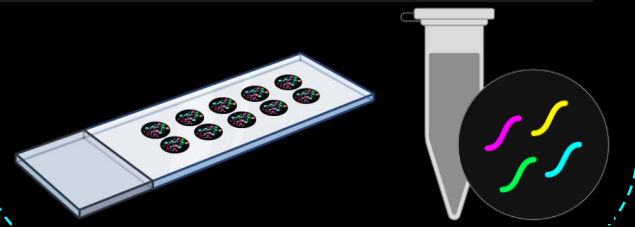
- ✓ **4-weeks+** earlier detection of variants of concern compared to NGS
- ✓ Dynamic panel modification with new probes within **2 weeks**
- ✓ Multiplex capability of **100+ targets** per sample – no sample splitting
- ✓ High and low abundance targets in **one reaction**
- ✓ NGS-grade **mutation frequency** quantification
- ✓ No need to rely on NGS for monitoring variants/mutations

Time for Action – Method Standardization is Key

- ✓ **Comparability** across European labs is essential
- ✓ Hyperplexed multi-pathogen panels are **now available**
- ✓ **Beta kits** are now ready to be delivered to your labs
- ✓ **Apply today** and get ahead with Hyperplex PCR™

Beta-kits

Beta applications open



Hyperplex PCR™

Powered by Nanopixels™

Revolutionizing Molecular Diagnostics

Contact:
Danai Nikou
Business Developer
danai@aplex.bio

Aplex BIO

Learn more at
www.aplex.bio

Extra slides

Sampling

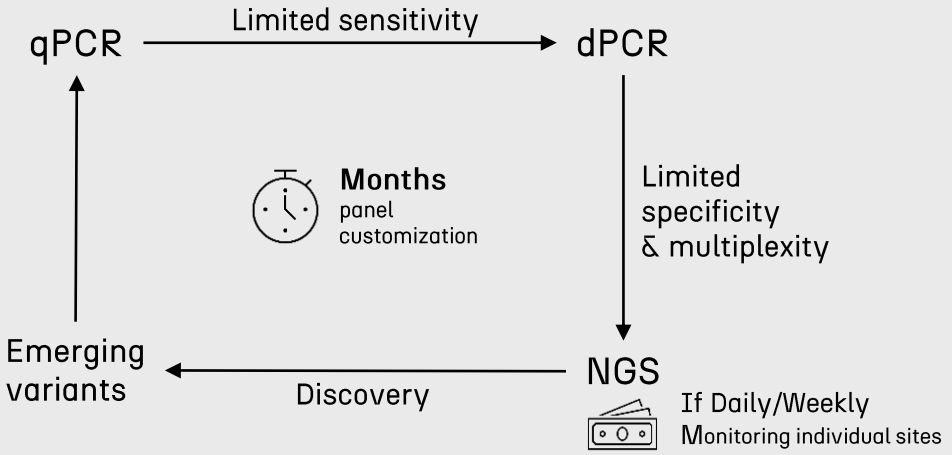
Population

Pathogens circulating in communities enter wastewater

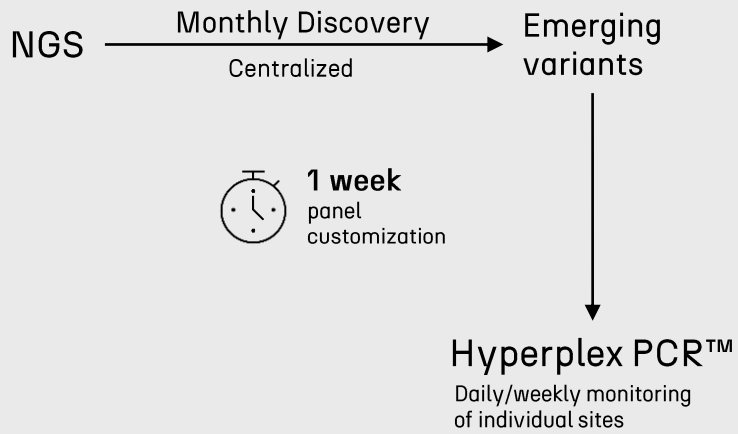
WW treatment plants

Sewage system gathers wastewater samples from communities

Discovery & Monitoring



Or



Time for Action

4+ Weeks

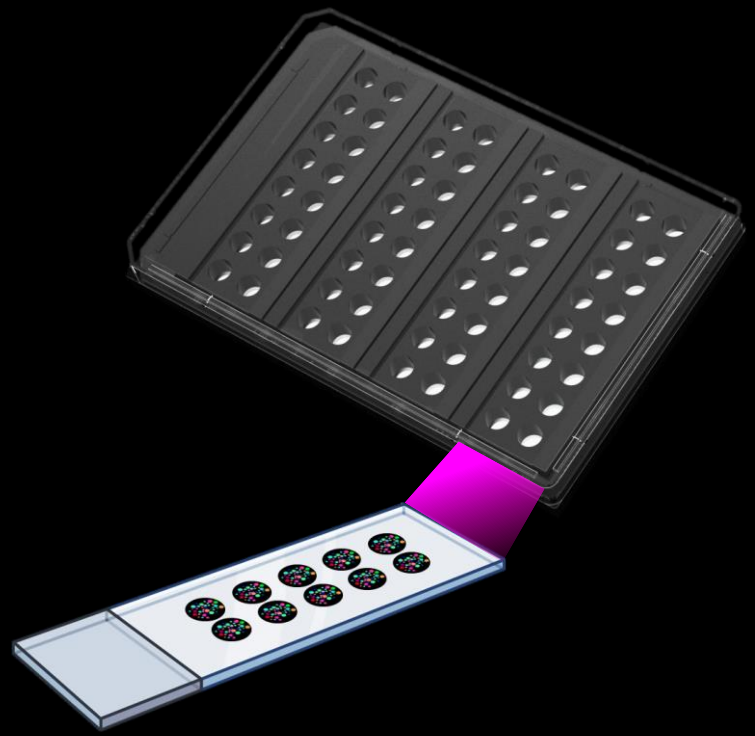
1 day

Comparable and Quantitative Results

Between sites

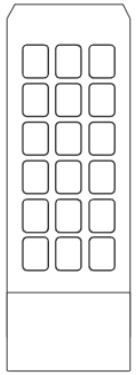
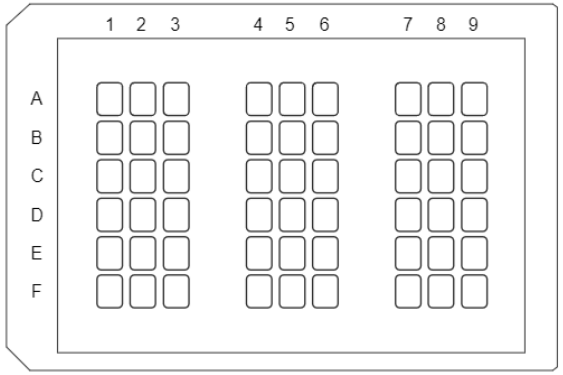
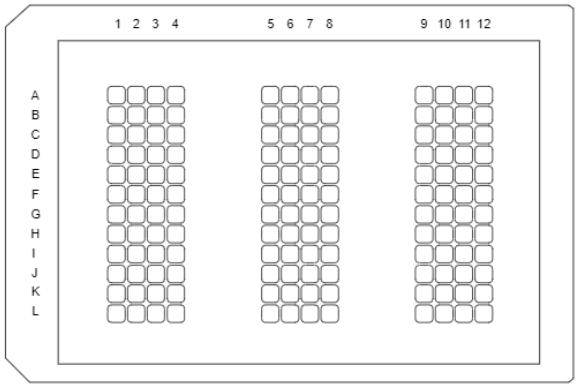
Throughput and read-out

SBS format
Up to 384 samples at once
Automated off the shelf



Feature	hpPCR
Multiplex/well	100+
Counts/well	Up to 1 000 000
Throughput/day	User/automation defined >128 samples
Sample type	Extracted DNA/RNA
Workflow	4-8 hr / <30 min hands-on
Software	Dedicated image analysis pipeline included
Instrumentation	PCR cycler Fluorescence microscope
Microscope	Objective: 20x/0.8 Channels: 4-6

High Throughput Enabled With Standard Format Consumables

	Current: Beta	SCALED	HIGH THROUGHPUT
			
Samples	18	54 (3x18)	144 (3x48)
Hyperplexing (per spot)	20	100	100
Or max multiplex (using all spots)	360	5 400	14 400

Consumables are designed to be compatible with standard plate formats as well as to facilitate flexibility with microscope slides