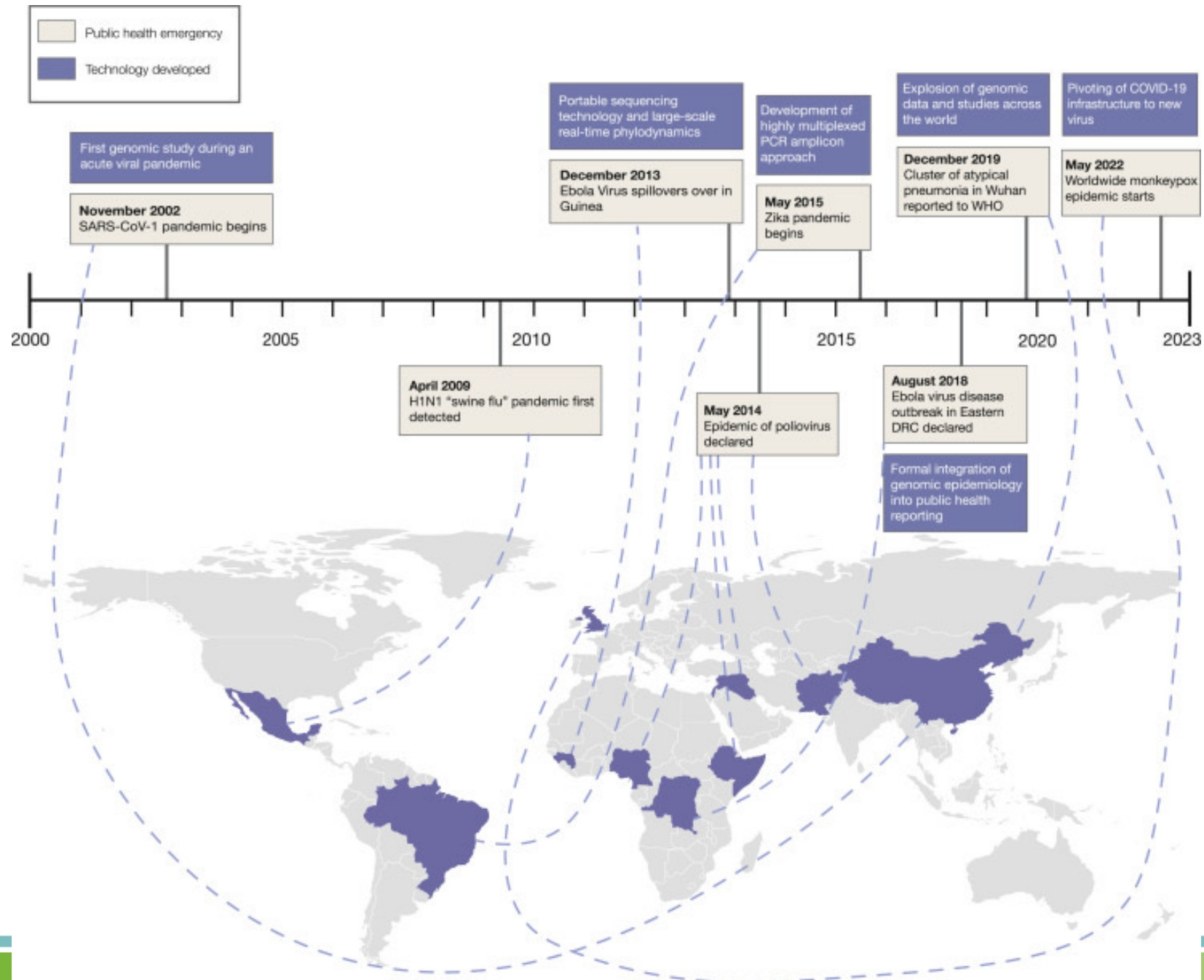




Mobile labs and field sequencing during outbreaks

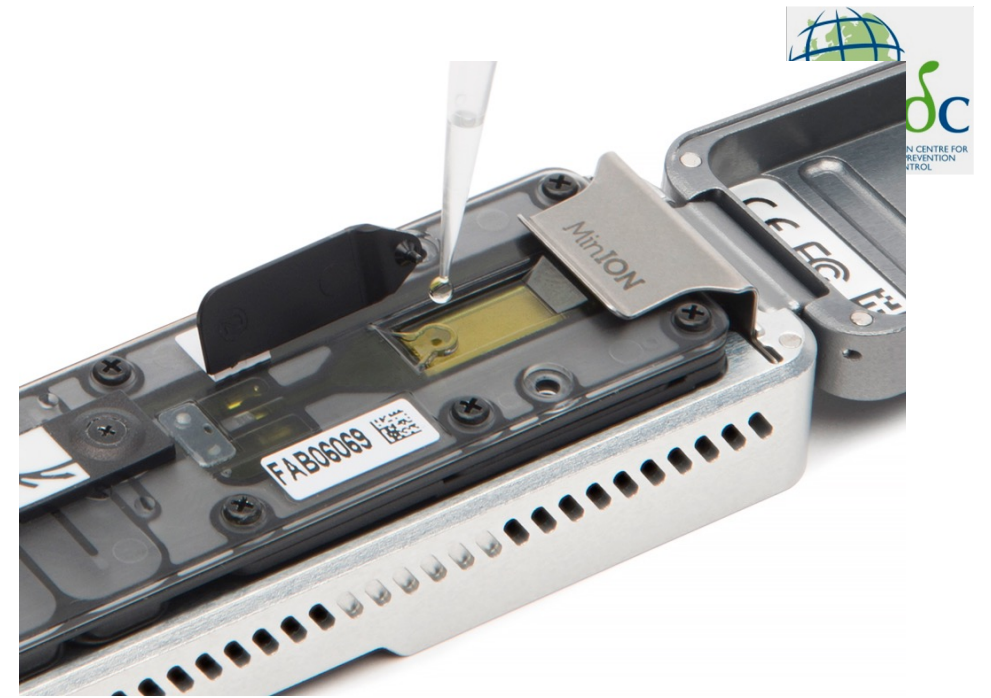
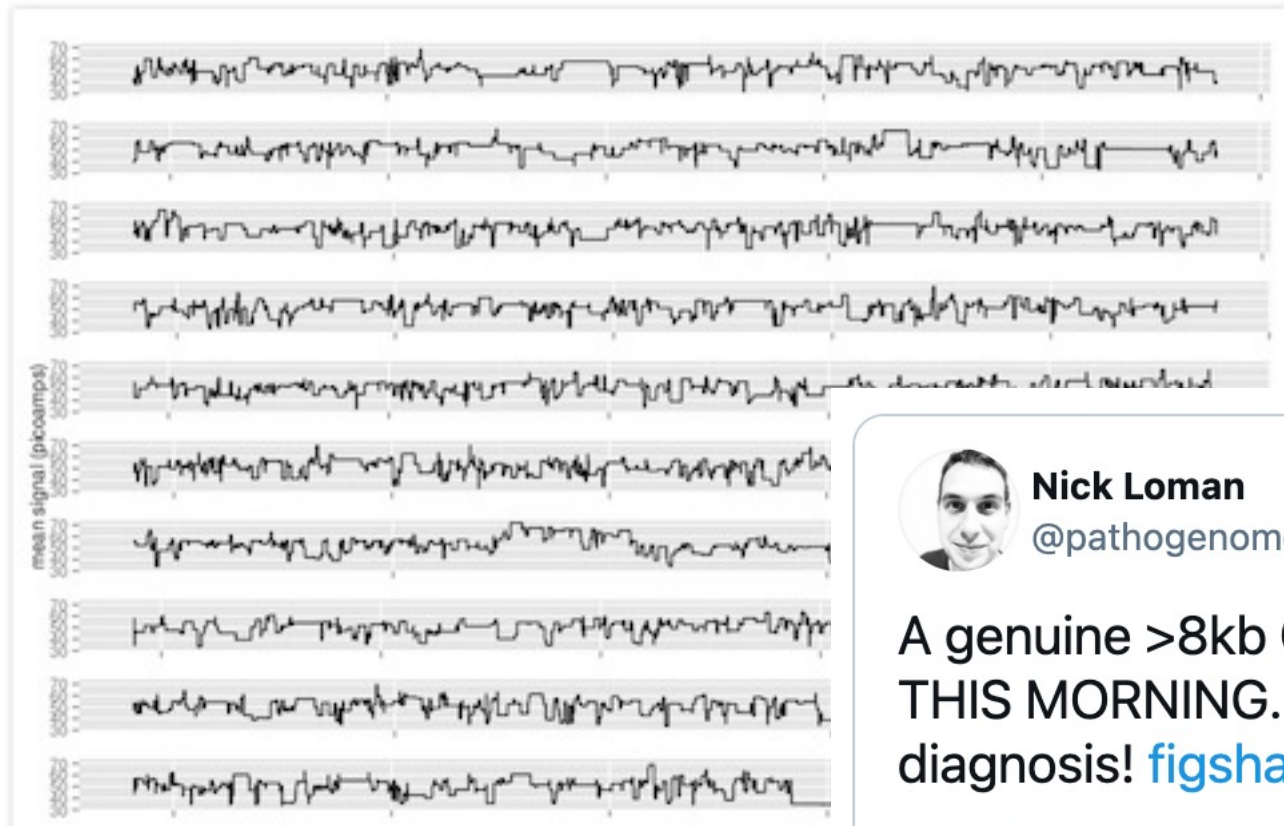
History and development of sequencing in the field

Joshua Quick, ARTIC network
15th April 2026





Early adopters



Nick Loman

@pathogenomenick



A genuine >8kb Oxford @nanopore read off our instrument THIS MORNING. And happily it's actually useful for diagnosis! figshare.com/articles/A_P_a...

7:40 AM · Jun 11, 2014



42

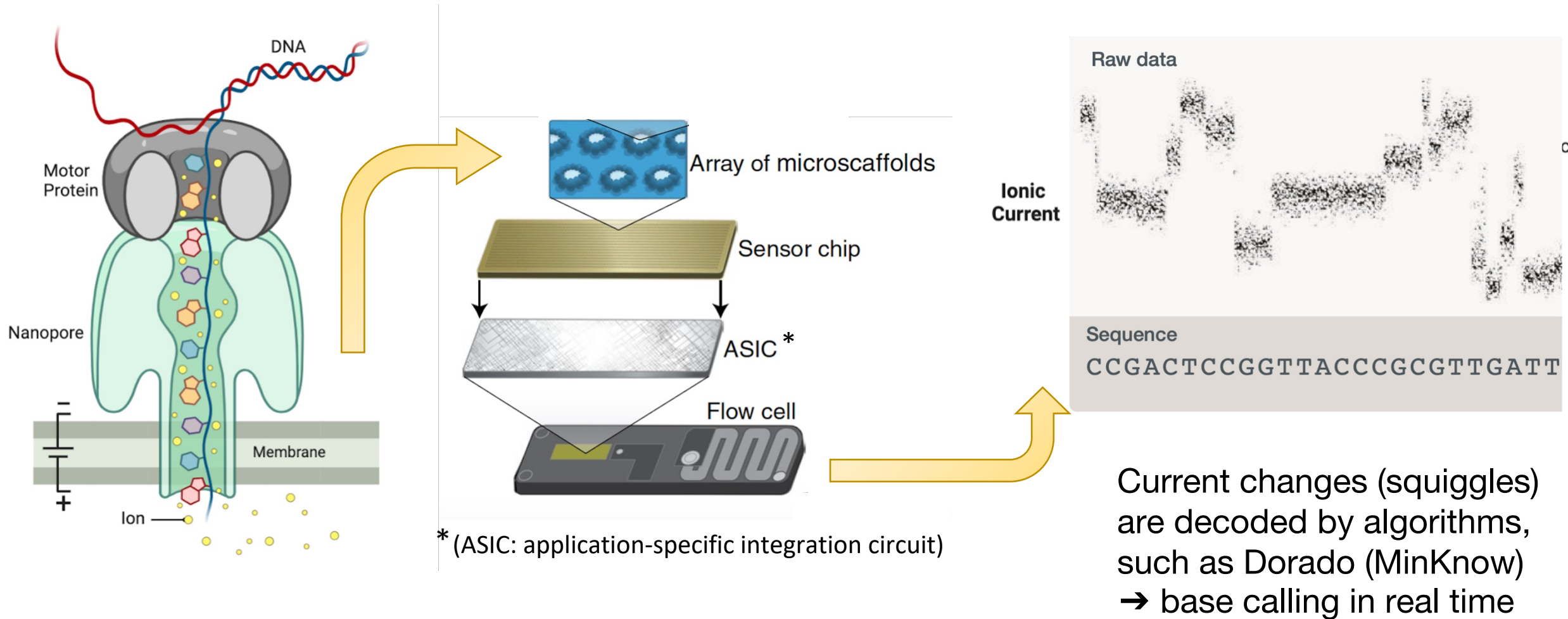


10

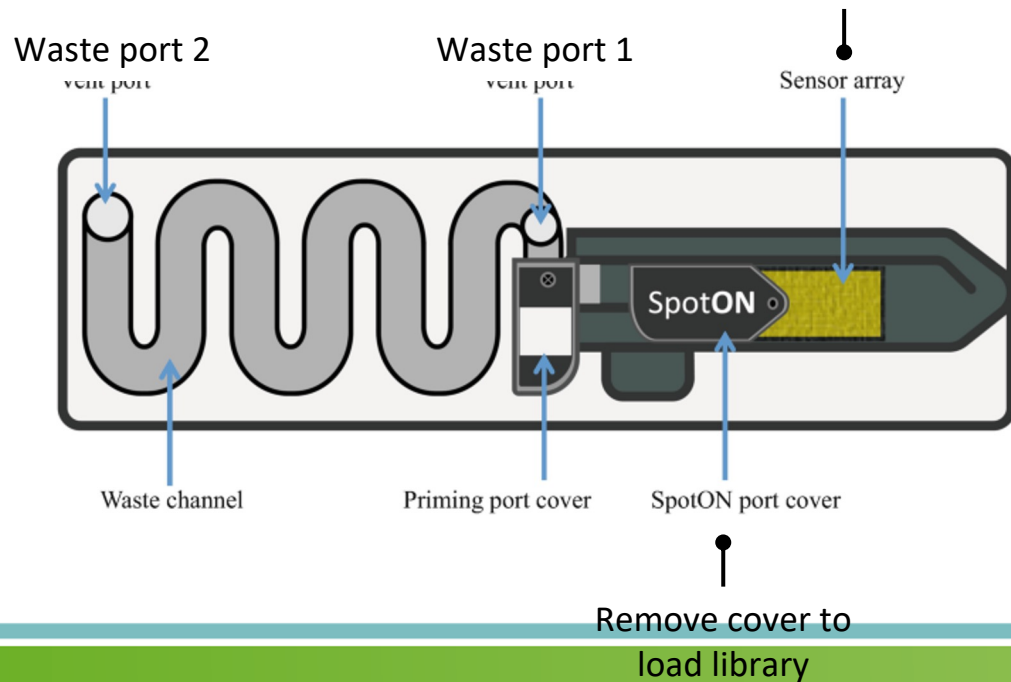


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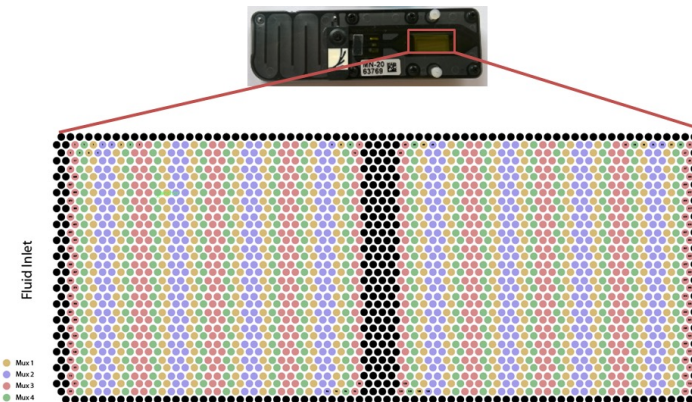
Principles of ONT sequencing



Flow cell at a glance

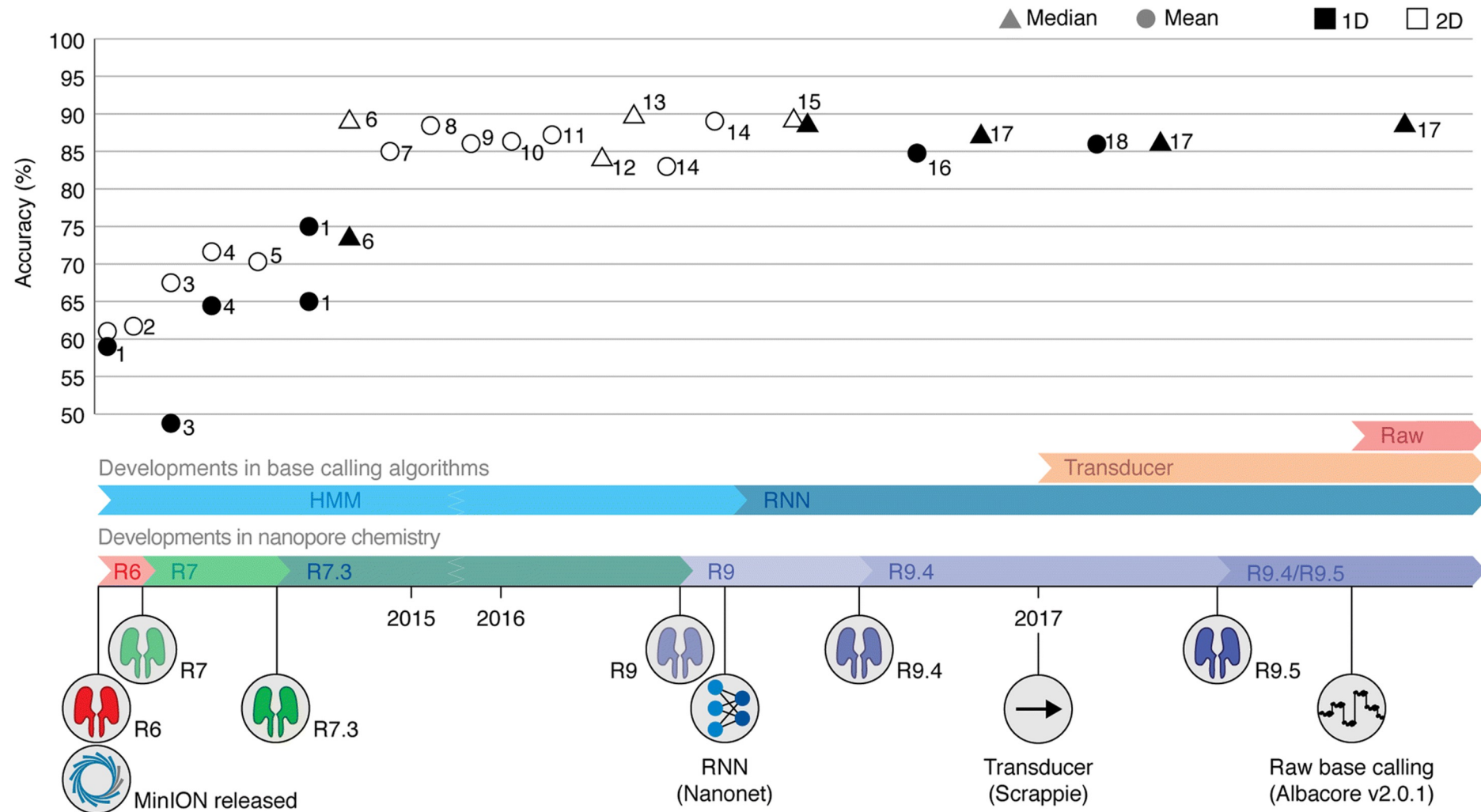


Warranty >800 active pores



2048 active well electrodes
512 channels → 4 channels

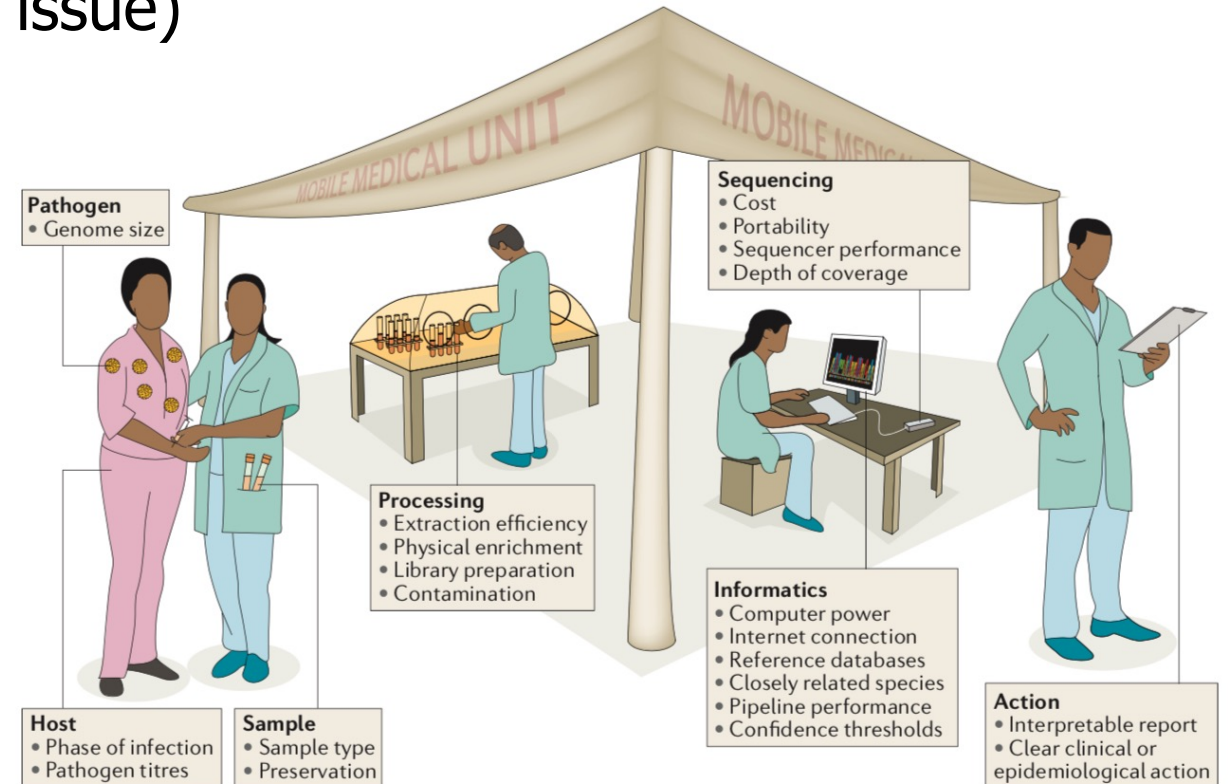




Rang FJ et al. From squiggle to basepair: computational approaches for improving nanopore sequencing read accuracy. *Genome Biol.* 2018 Jul 13;19(1):90.

Why sequence in the field?

- Sample integrity
- Shipping dangerous samples
- Nagoya protocol (soon to cover data)
- Faster results
- Build local capacity (logistics still an issue)

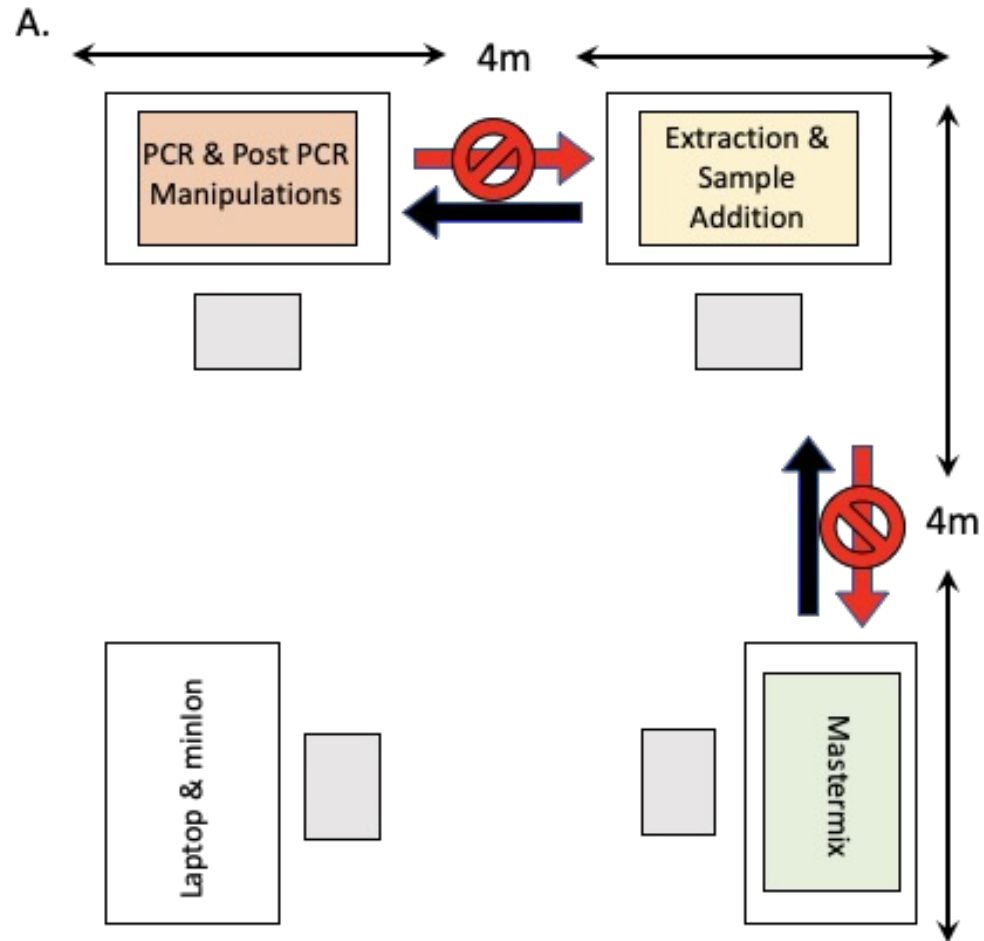




<https://twitter.com/igoodfel/>



Portable Sequencing Lab



B.

INRB Kinshasa

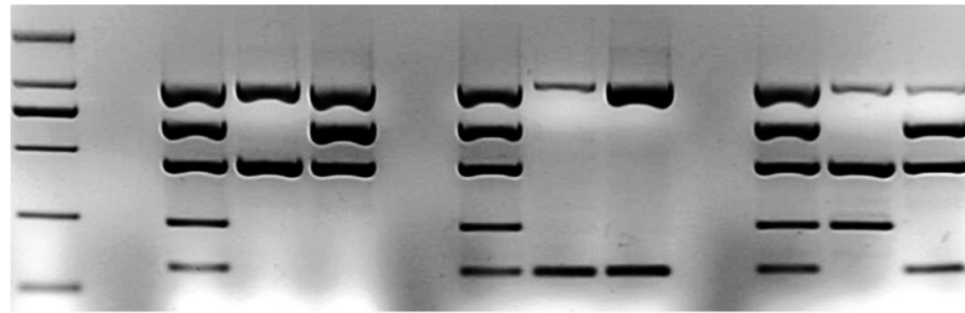


Goma

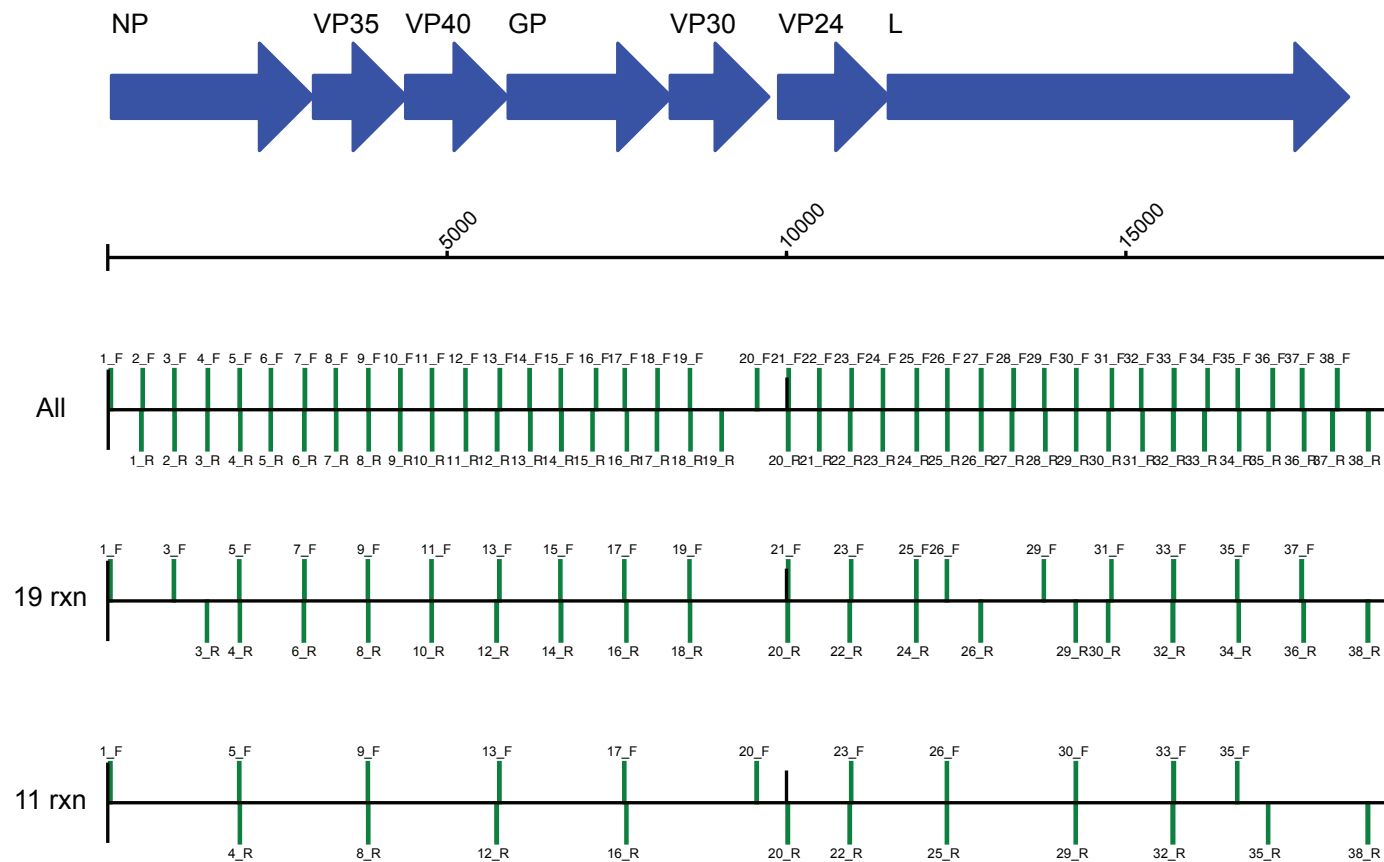


↑
JJ Muyembe





A.

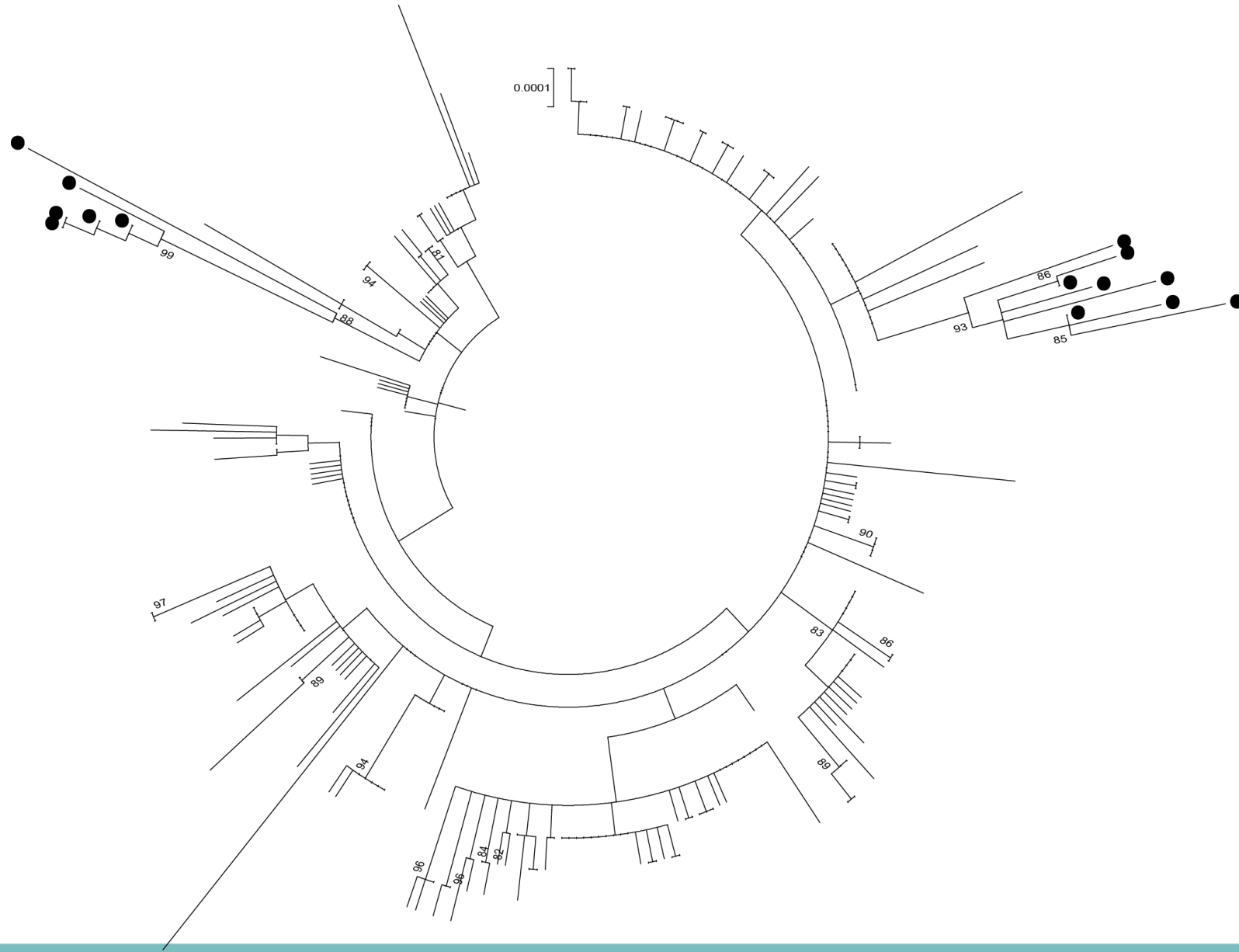


B. 19 reactions

Forward	Reverse	Length
1_F	3_R	1426
3_F	4_R	973
5_F	6_R	952
7_F	8_R	941
9_F	10_R	940
11_F	12_R	958
13_F	14_R	906
15_F	16_R	974
17_F	18_R	969
19_F	20_R	1445
21_F	22_R	906
23_F	24_R	958
25_F	26_R	947
26_F	29_R	1898
29_F	30_R	946
31_F	32_R	901
33_F	34_R	963
35_F	36_R	977
37_F	38_R	975

C. 11 reactions

Forward	Reverse	Length
1_F	4_R	1911
5_F	8_R	1901
9_F	12_R	1895
13_F	16_R	1874
17_F	20_R	2406
20_F	22_R	1371
23_F	25_R	1410
26_F	29_R	1898
30_F	32_R	1427
33_F	35_R	1396
35_F	38_R	1921





Lauren Cowley @LaurenCowley4 · Jun 5

#Ebola **#MinION** sequencing right in the field, all in a days work



40



35



Sophie Duraffour

Lauren Cowley

Joseph Bore

Raymond Koundouno

Antonio Mazarelli

Liana Kafetzopoulou

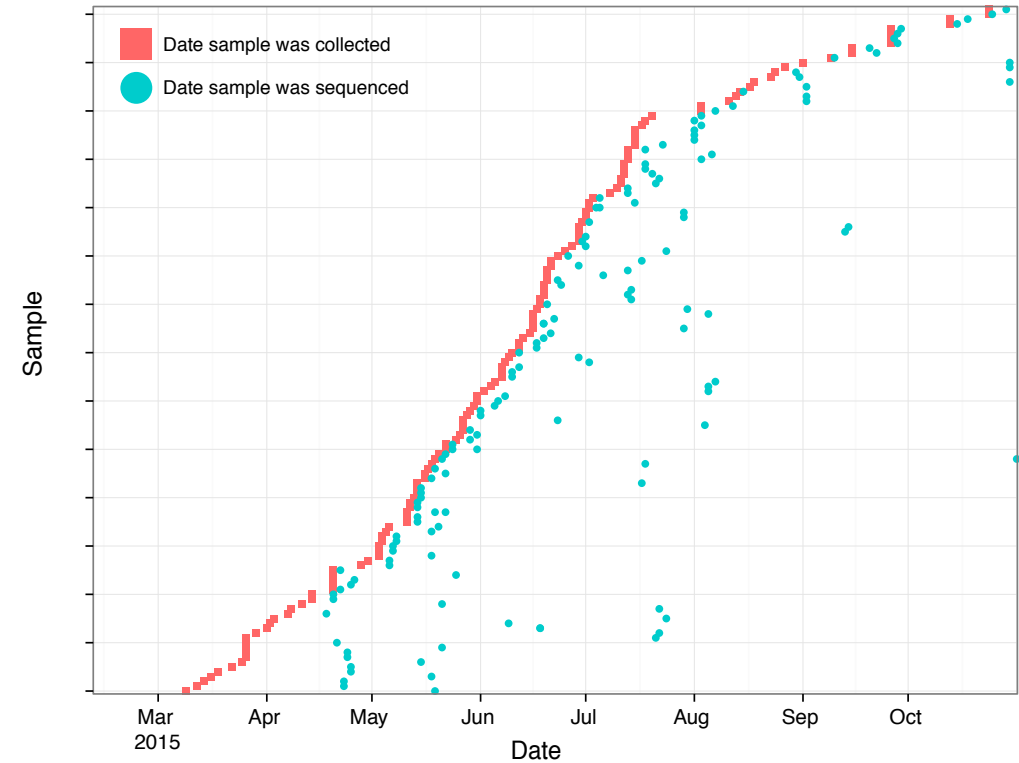
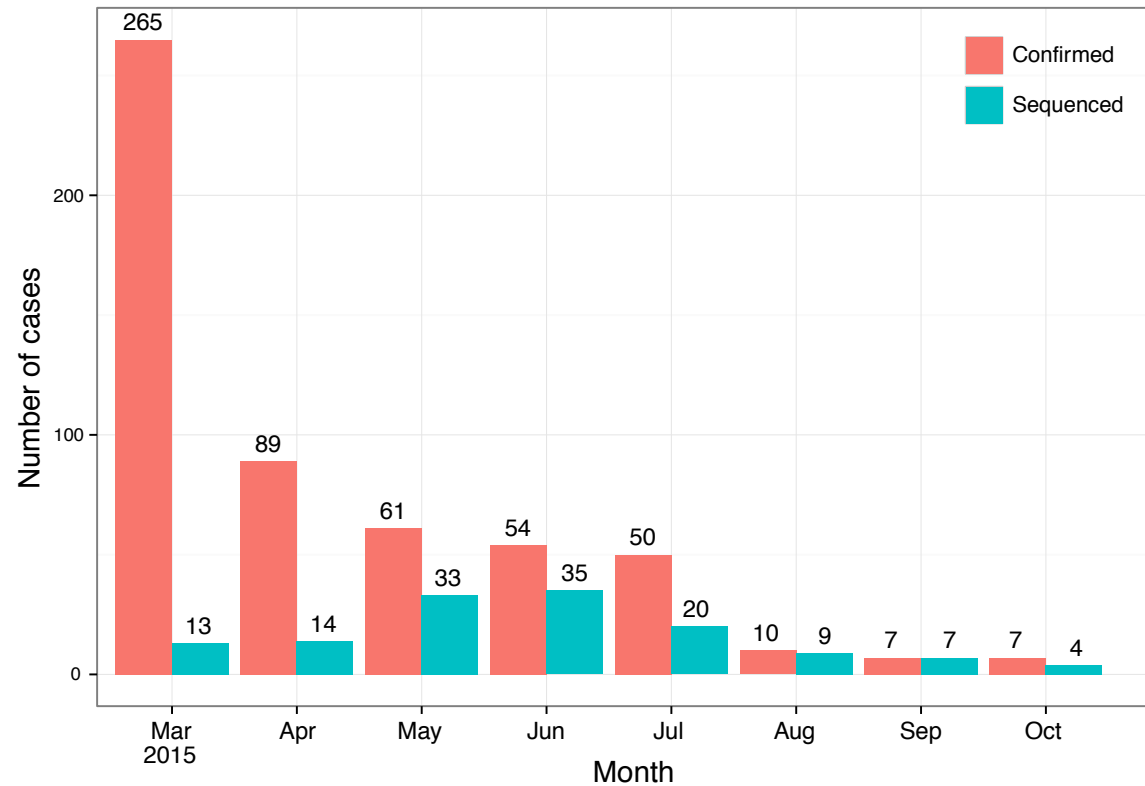


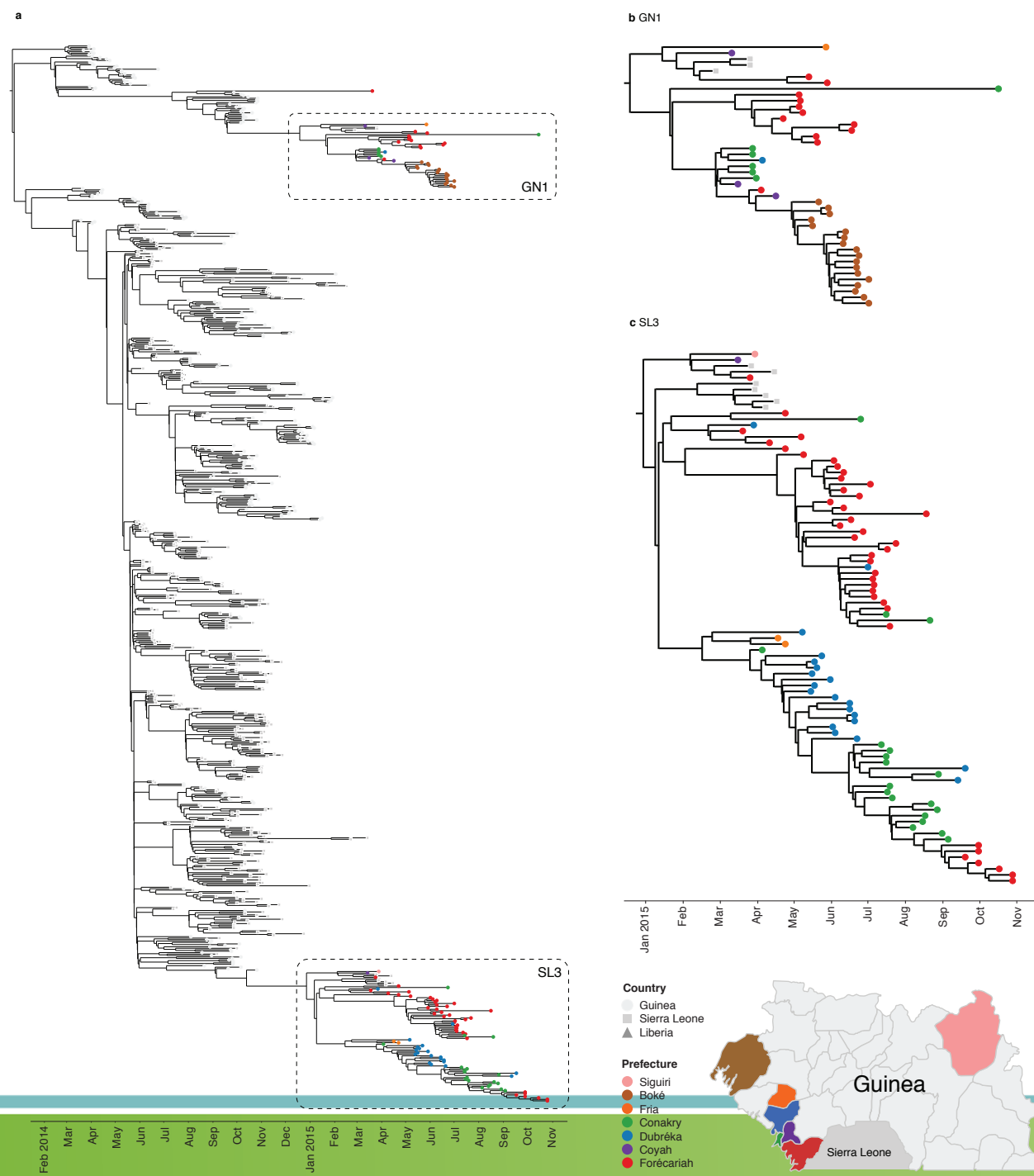






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Alert! There's a dangerous new viral outbreak: Zika conspiracy theories



Cancer treatment development to be speeded up by new London research hub

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Scientific ignorance about Zika parallels Aids crisis in 1980s, say Brazilian experts

Zika virus: pesticides are not a long-term solution says leading entomologist

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The laboratory in a suitcase

How genome sequencing can be quickly deployed to monitor outbreaks

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SCIENCE HEALTH REPORT

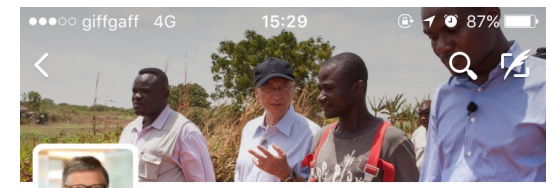
This 'lab in a suitcase' helped track Ebola cases last year

By **Russell Brandom** on February 3, 2016 01:00 pm [Email](#) [@russellbrandom](#)



PART OF THIS STORYSTREAM

49 UPDATES TO
Tracking the deadly ebola outbreak in West Africa



Bill Gates @BillGates

Sharing things I'm learning through my foundation work and other interests...

Seattle, WA [gatesnotes.com](#)

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Bill Gates @BillGates 1h

From Ebola to Zika, this "lab in a suitcase" provides crucial data for outbreaks: [b-gat.es/1XIKQkZ](#) via @verge



Zoophilic mosquitoes

<i>Ae. dalzieli</i>	<i>Ae. tarsalis</i>	<i>Ae. taeniarostris</i>
<i>Ae. fowleri</i>	<i>Ae. vitattus</i>	<i>Ae. tarsalis</i>
<i>Ae. furcifer</i>	<i>Ae. africanus</i>	<i>Ae. vitattus</i>
<i>Ae. luteocephalus</i>	<i>Ae. flavicollis</i>	<i>An. gambiae</i>
<i>Ae. metallicus</i>	<i>Ae. furcifer</i>	<i>Mansonia uniformis</i>
<i>Ae. minutus</i>	<i>Ae. grahami</i>	<i>Eratmapodites</i>
<i>Ae. neoaffricanus</i>	<i>Ae. opok</i>	



Vector-borne transmission



**Sylvatic
cycle**



Anthropophilic mosquitoes

Ae. aegypti
Ae. albopictus
Ae. hensilii



**Urban
cycle**



Amniotic fluid
Semen
Urine
Saliva
Breast milk
Blood

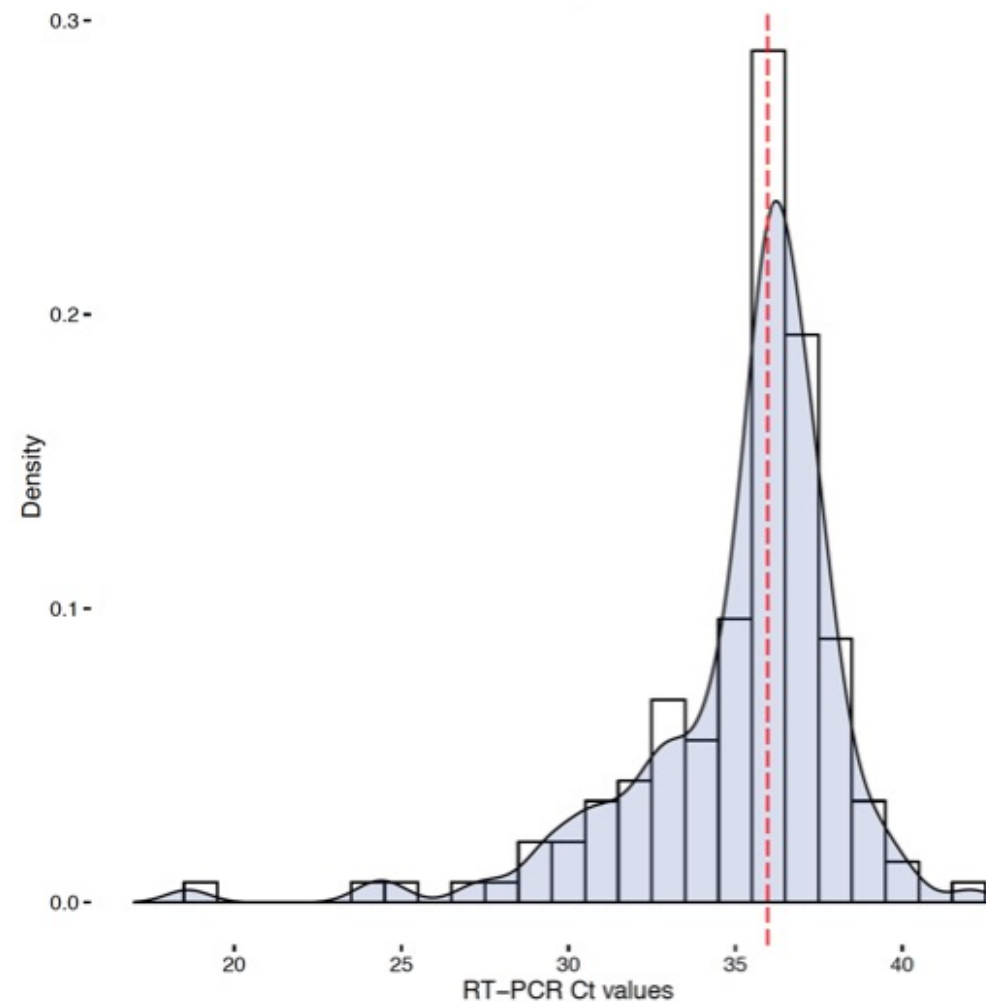


**Non-Vector-
borne
transmission**



Technical challenge: High Ct

- Direct RNA
- RNA-Seq (w/ DNase)
- Bait capture
- Amplicon
- qRT-PCR



dRNA

RNA-Seq

Bait capture

Amplicon

qRT-PCR

ZiBRA project data

primalscheme

primer panels for multiplex PCR



Design a new scheme

Reset defaults



FASTA file...

nCoV-2019.reference.fas...

Options



High-GC mode



Pinned



Use the [standard protocol](#) for these settings.

Amplicon size

400

Min/max will be set at 5% either side of target.

Scheme name

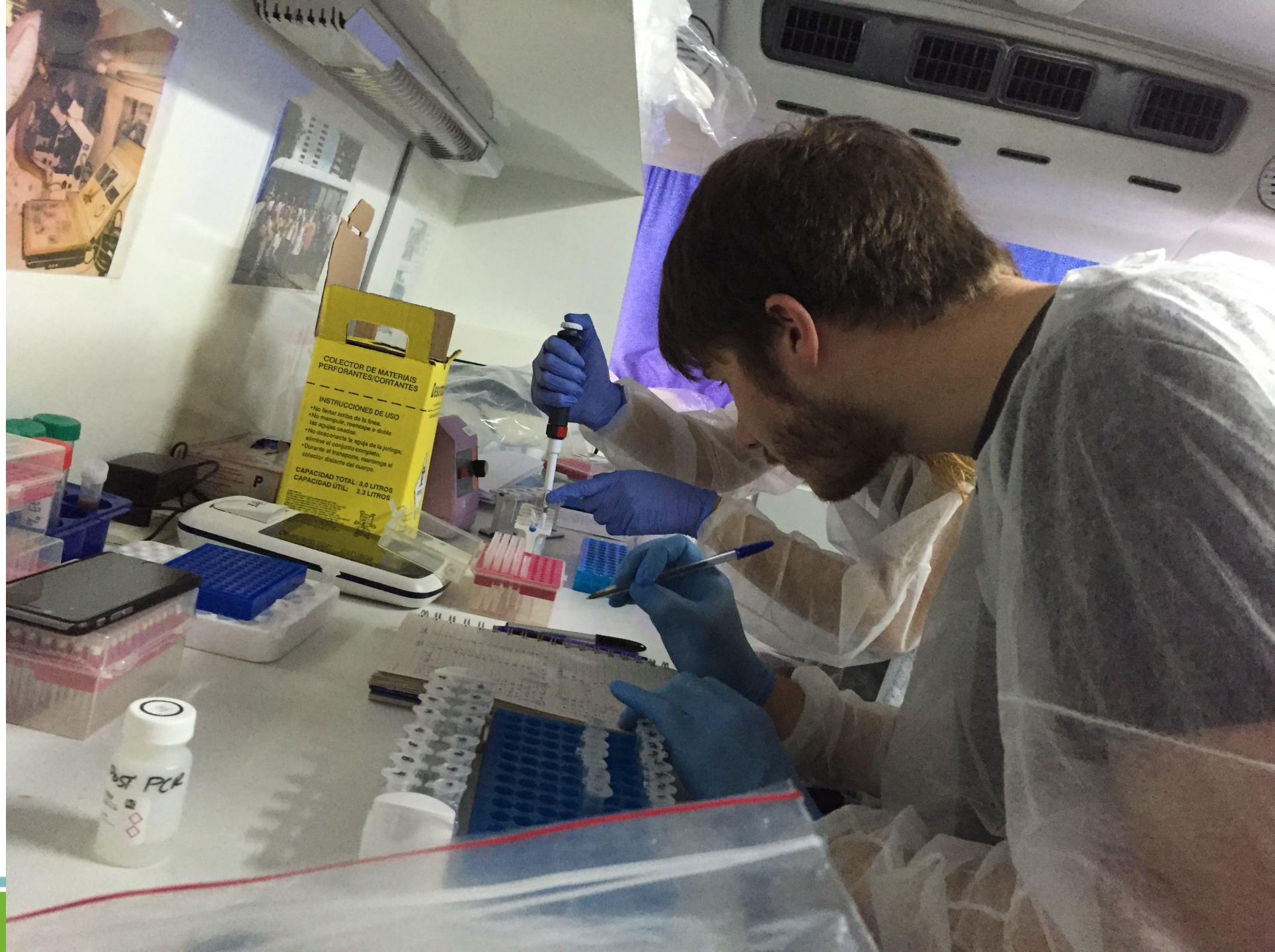
nCoV-400

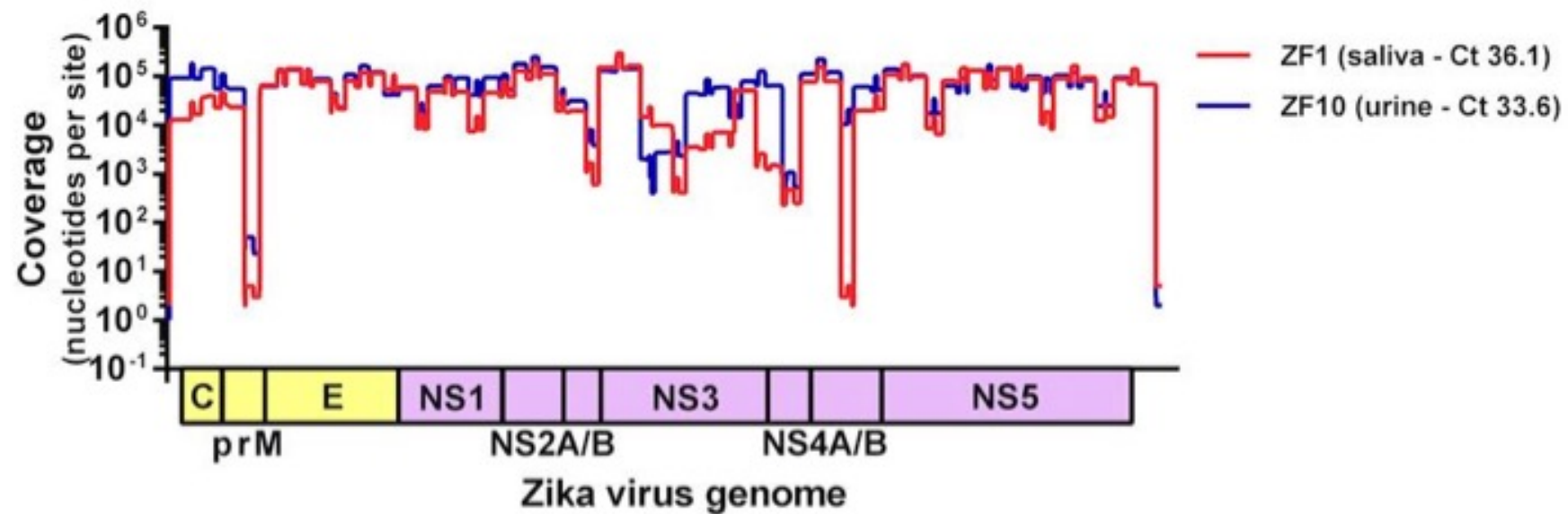
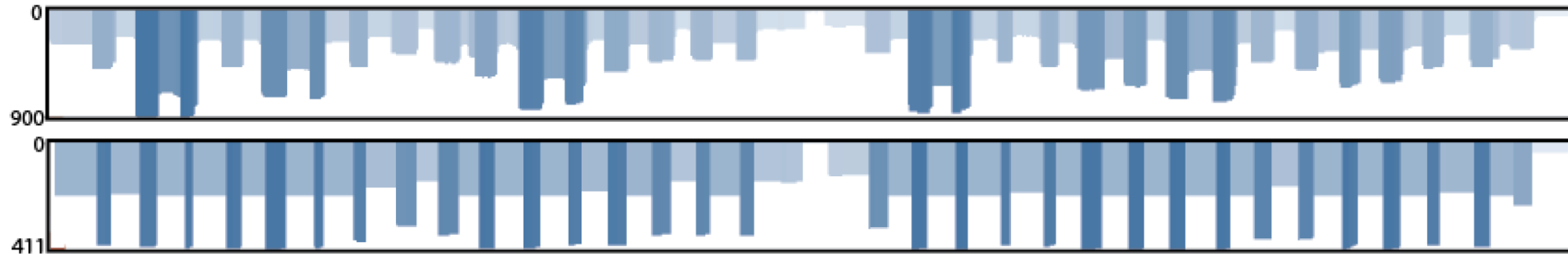
A short name/prefix for your scheme, no spaces.



Design scheme

View [demo inputs](#)

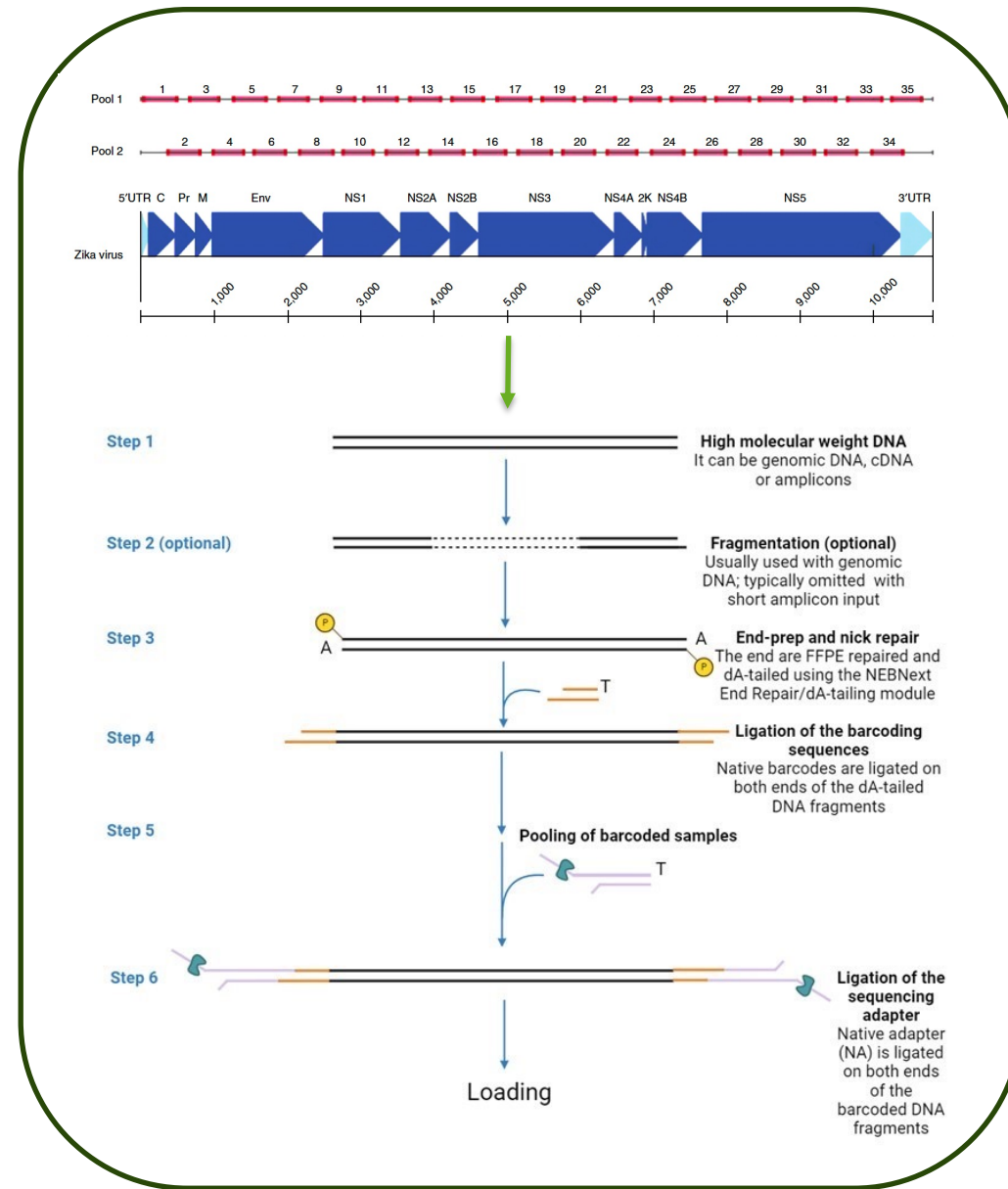




Two Florida samples Ct 33.6 and 36.1 sequenced in the Andersen Lab

Current ONT workflow

Current ARTIC workflow uses
barcode ligation
Very low rate of barcode
swapping/misidentification
Limited room for further
optimisation



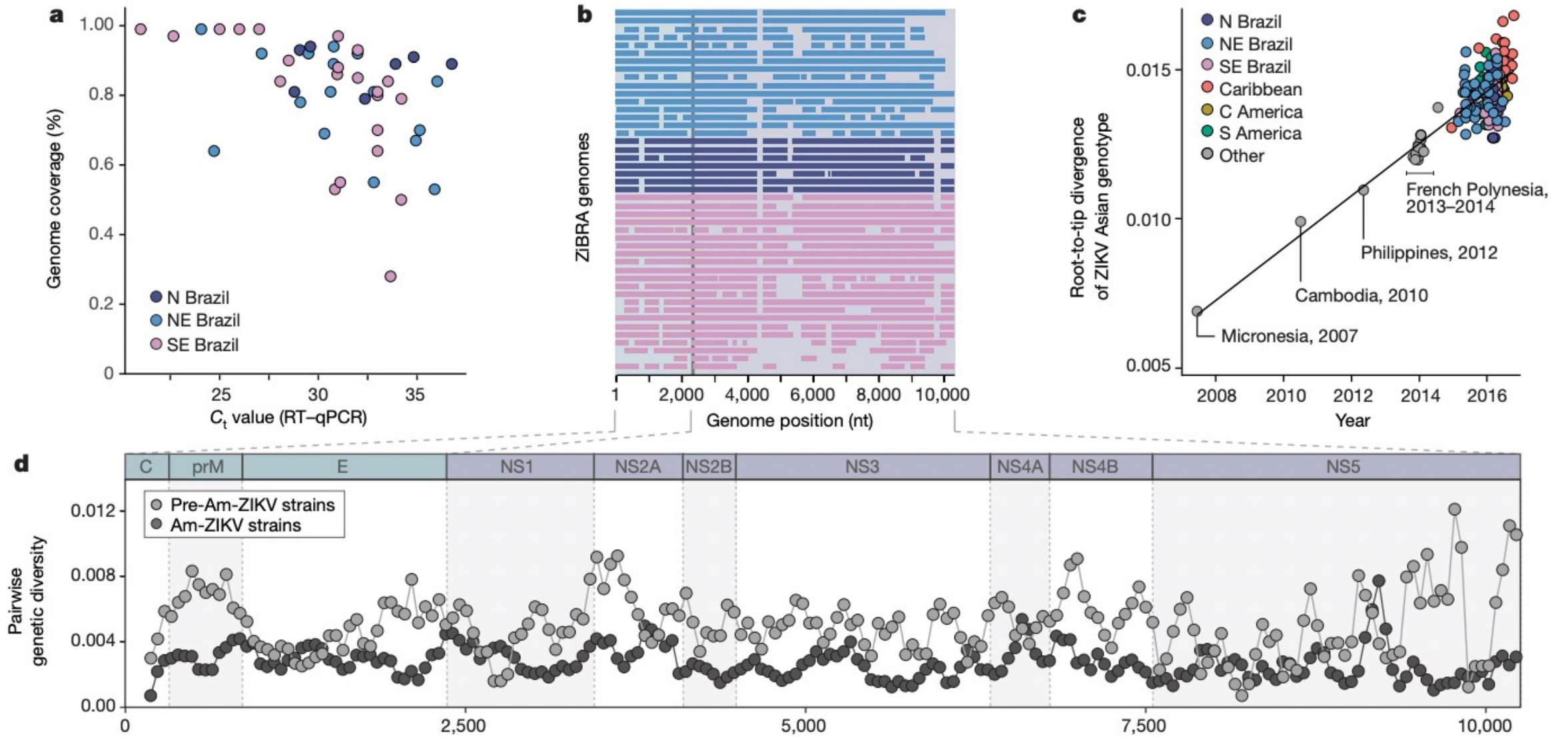
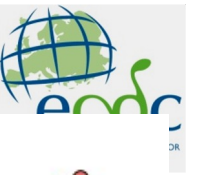
Multiplex PCR method for MinION and Illumina sequencing of Zika and other virus genomes directly from clinical samples

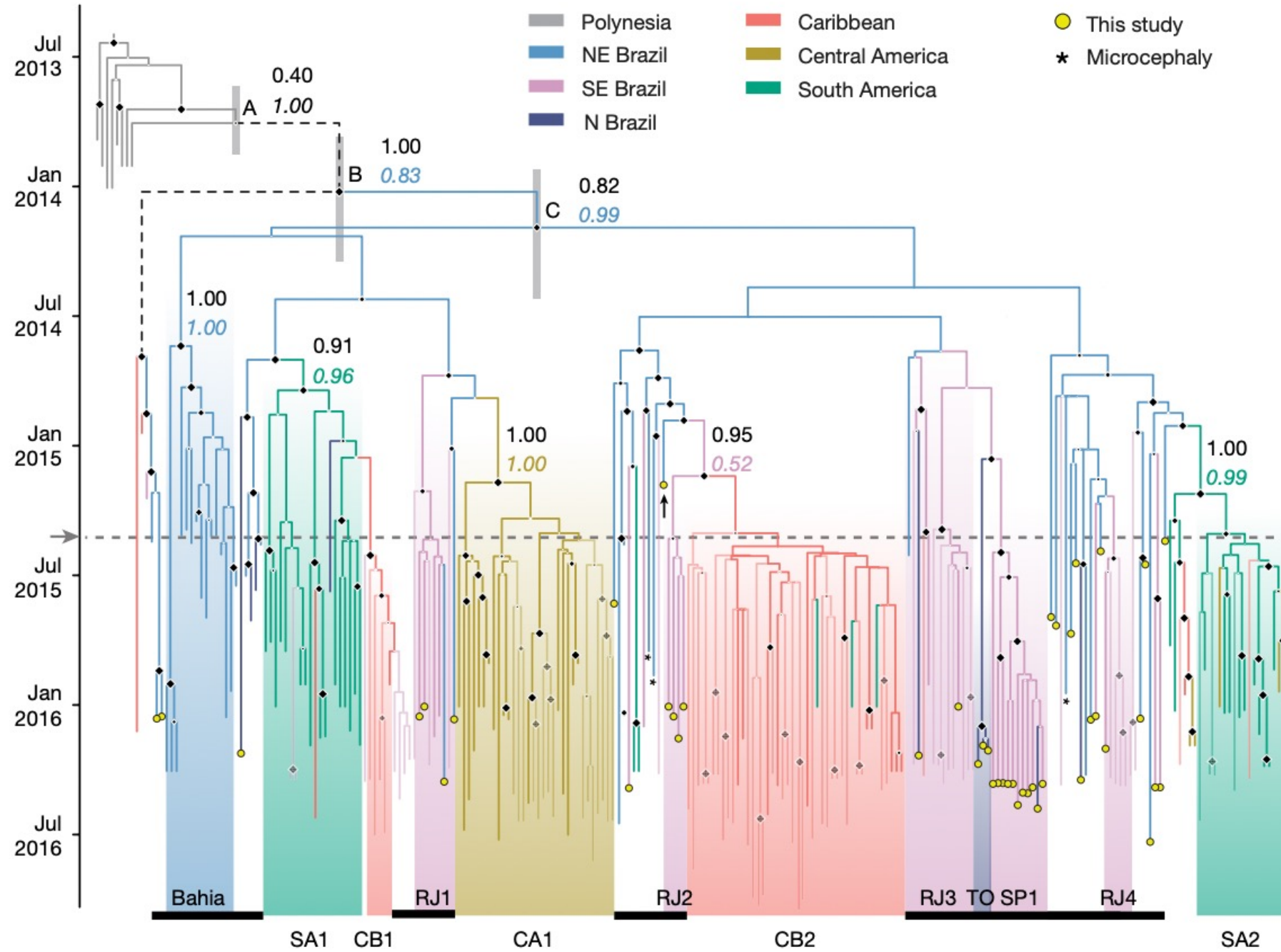
Joshua Quick¹, Nathan D Grubaugh² , Steven T Pullan³, Ingra M Claro⁴, Andrew D Smith¹, Karthik Gangavarapu², Glenn Oliveira⁵, Refugio Robles-Sikisaka², Thomas F Rogers^{2,6}, Nathan A Beutler², Dennis R Burton², Lia Laura Lewis-Ximenez⁷, Jaqueline Goes de Jesus⁸, Marta Giovanetti^{8,9}, Sarah C Hill¹⁰, Allison Black^{11,12} , Trevor Bedford¹¹, Miles W Carroll^{3,13}, Marcio Nunes¹⁴, Luiz Carlos Alcantara Jr.⁸ , Ester C Sabino⁴, Sally A Baylis¹⁵, Nuno R Faria¹⁰, Matthew Loose¹⁶, Jared T Simpson¹⁷, Oliver G Pybus¹⁰, Kristian G Andersen^{2,5} & Nicholas J Loman¹

¹Institute of Microbiology and Infection, School of Biosciences, University of Birmingham, Birmingham, UK. ²The Scripps Research Institute, La Jolla, California, USA. ³Public Health England, National Infection Service, Porton Down, Salisbury, UK. ⁴Department of Infectious Disease and Institute of Tropical Medicine, University of São Paulo, São Paulo, Brazil. ⁵Scripps Translational Science Institute, La Jolla, California, USA. ⁶Massachusetts General Hospital, Boston, Massachusetts, USA. ⁷Instituto Oswaldo Cruz, Fundação Oswaldo Cruz, Rio de Janeiro, Brazil. ⁸Fundação Oswaldo Cruz (FIOCRUZ), Salvador, Brazil. ⁹University of Rome, Tor Vergata, Italy. ¹⁰Department of Zoology, University of Oxford, Oxford, UK. ¹¹Vaccine and Infectious Disease Division, Fred Hutchinson Cancer Research Center, Seattle, Washington, USA. ¹²Department of Epidemiology, University of Washington, Seattle, Washington, USA. ¹³University of Southampton, South General Hospital, Southampton, UK. ¹⁴Instituto Evandro Chagas, Belem, Brazil. ¹⁵Paul-Ehrlich-Institut, Langen, Germany. ¹⁶DeepSeq, School of Life Sciences, University of Nottingham, Nottingham, UK. ¹⁷OICR, Toronto, Canada. Correspondence should be addressed to N.J.L. (n.j.loman@bham.ac.uk).

Published online 24 May 2017; doi:10.1038/nprot.2017.066

Published online 24 May 2017; doi:10.1038/nprot.2017.066







Severe acute respiratory syndrome coronavirus 2 isolate Wuhan-Hu-1, complete genome

GenBank: MN908947.3

[FASTA](#) [Graphics](#)

[Go to:](#) ☐

LOCUS	MN908947	29903 bp ss-RNA	linear	VRL	18-MAR-2020
DEFINITION	Severe acute respiratory syndrome coronavirus 2 isolate Wuhan-Hu-1, complete genome.				
ACCESSION	MN908947				
VERSION	MN908947.3				
KEYWORDS	.				
SOURCE	Severe acute respiratory syndrome coronavirus 2 (SARS-CoV-2)				
ORGANISM	Severe acute respiratory syndrome coronavirus 2 Viruses; Riboviria; Orthornavirae; Pisuviricota; Pisoniviricetes; Nidovirales; Cornidovirineae; Coronaviridae; Orthocoronavirinae; Betacoronavirus; Sarbecovirus.				
REFERENCE	1 (bases 1 to 29903)				
AUTHORS	Wu,F., Zhao,S., Yu,B., Chen,Y.M., Wang,W., Song,Z.G., Hu,Y., Tao,Z.W., Tian,J.H., Pei,Y.Y., Yuan,M.L., Zhang,Y.L., Dai,F.H., Liu,Y., Wang,Q.M., Zheng,J.J., Xu,L., Holmes,E.C. and Zhang,Y.Z.				
TITLE	A new coronavirus associated with human respiratory disease in China				
JOURNAL	Nature 579 (7798), 265-269 (2020)				
PUBMED	32015508				
REFERENCE	2 (bases 1 to 29903)				
AUTHORS	Wu,F., Zhao,S., Yu,B., Chen,Y.-M., Wang,W., Hu,Y., Song,Z.-G., Tao,Z.-W., Tian,J.-H., Pei,Y.-Y., Yuan,M.L., Zhang,Y.-L., Dai,F.-H., Liu,Y., Wang,Q.-M., Zheng,J.-J., Xu,L., Holmes,E.C. and Zhang,Y.-Z.				
TITLE	Direct Submission				
JOURNAL	Submitted (05-JAN-2020) Shanghai Public Health Clinical Center & School of Public Health, Fudan University, Shanghai, China				
COMMENT	On Jan 17, 2020 this sequence version replaced MN908947.2 .				



Version 1 ☐

Jan 22, 2020

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nCoV-2019 sequencing protocol V.1 ☐
 Forked from [Ebola virus sequencing protocol](#)

Spanish Journal of Legal Medicine

Josh Quick¹
¹University of Birmingham

18 *Works for me* [dx.doi.org/10.17504/protocols.io.bbmuik6w](https://doi.org/10.17504/protocols.io.bbmuik6w)

ARTIC **Coronavirus Method Development Community** 1 more workspace

Josh Quick

SUBMIT TO PLOS ONE

Steps Forks Metadata Metrics

ABSTRACT
ARTIC amplicon sequencing protocol for MinION for nCoV-2019



ARTIC amplicon sequencing protocol for MinION for nCoV-2019

ABSTRACT

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NEBNext[®] ARTIC

[SARS-CoV-2. Don't let up. We'll help.](#)

Product Listing

Interested in trying NEBNext AR

Especially with the ongoing emerg health, there is an increasing need based on the original work of the A sequencing protocol v2 (GunIt)).

The ARTIC method is a multiplexed options are compatible with Illumin sequencing generate library inserts

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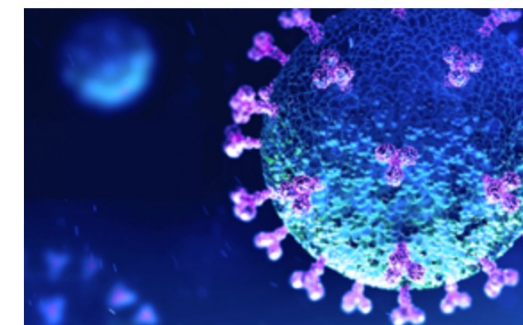
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COVID-19

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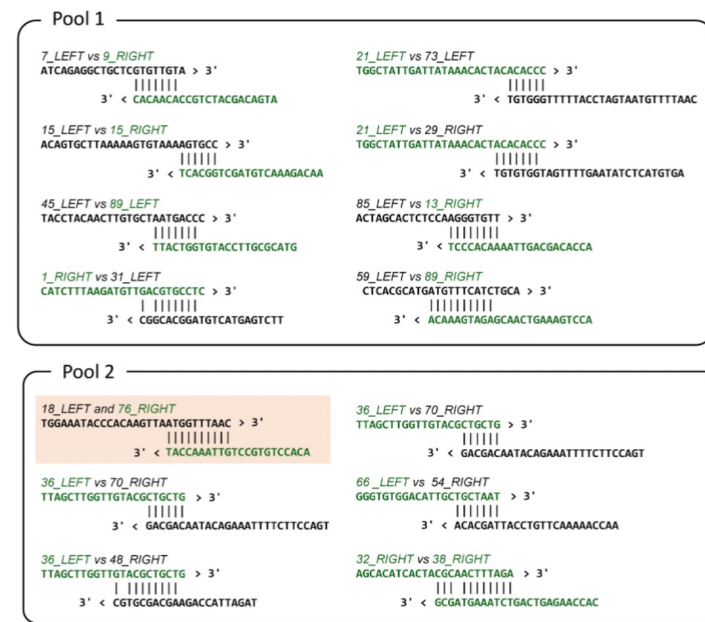
**Coronavirus
Characterization
Products**

EXPEDITING YOUR COVID-19 RESPONSE, VACCINE

As a global life science supplier, we are here to help in the response for detection and characterization to production, we've consor

primalscheme3

68k jobs, 18M Covid19 genomes
Developments targeting more diversity,
multiple reference panels, ML interactions
checking
Uses MSA alignment as input
Use more primers without increasing
interactions aiming for megaplex PCR



Most common interactions in ARTIC V3 scheme (Itokawa et al.)

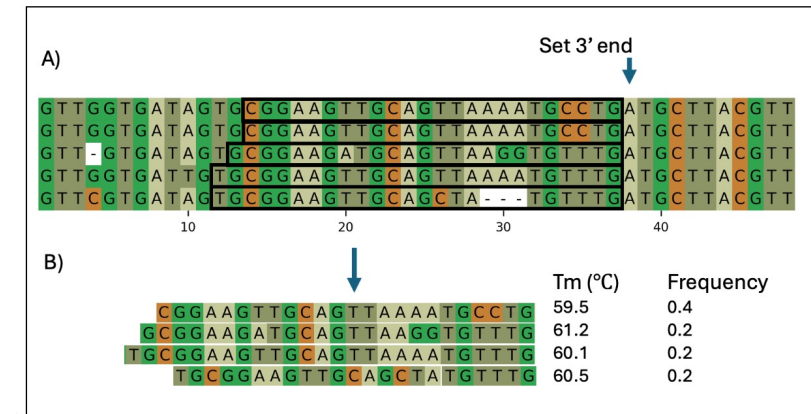


Figure 4. An overview of the digestion algorithm which generates the discrete primer clouds. **A)** shows an example input alignment. After establishing the 3' end, primers are extended until reaching the target Tm range (black boxes). **B)** shows the resulting primer cloud, Tm of each primer, and the frequency each primer occurs in the original alignment.

Pan-Measles scheme

Able to amplify any genotype of
Measles with a single scheme
Up to 12% diversity between genomes
High sensitivity from short amplicons
Can be used for wastewater
surveillance

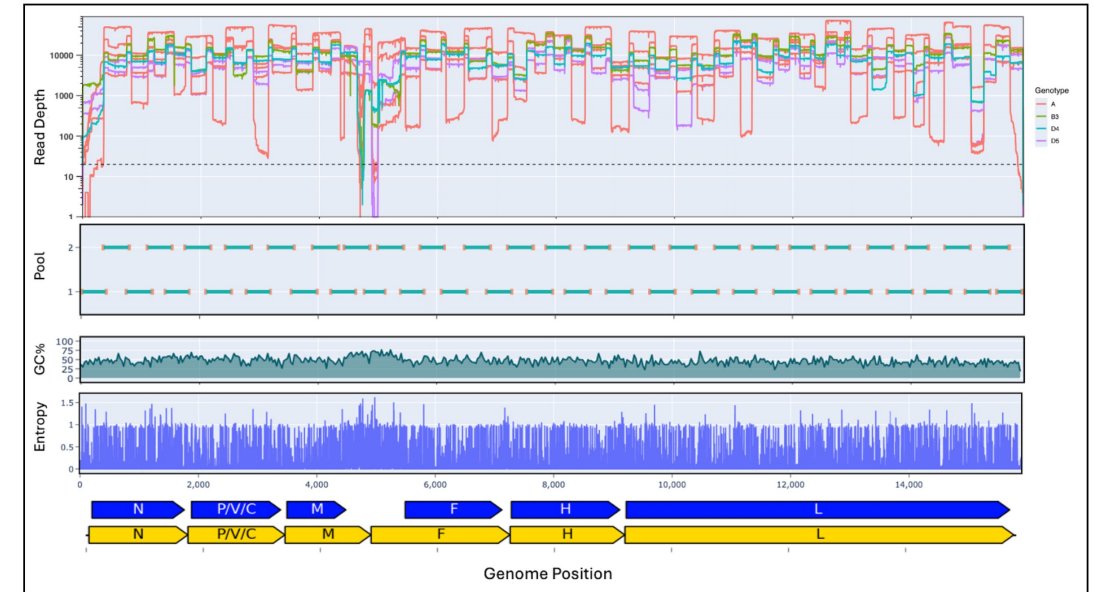
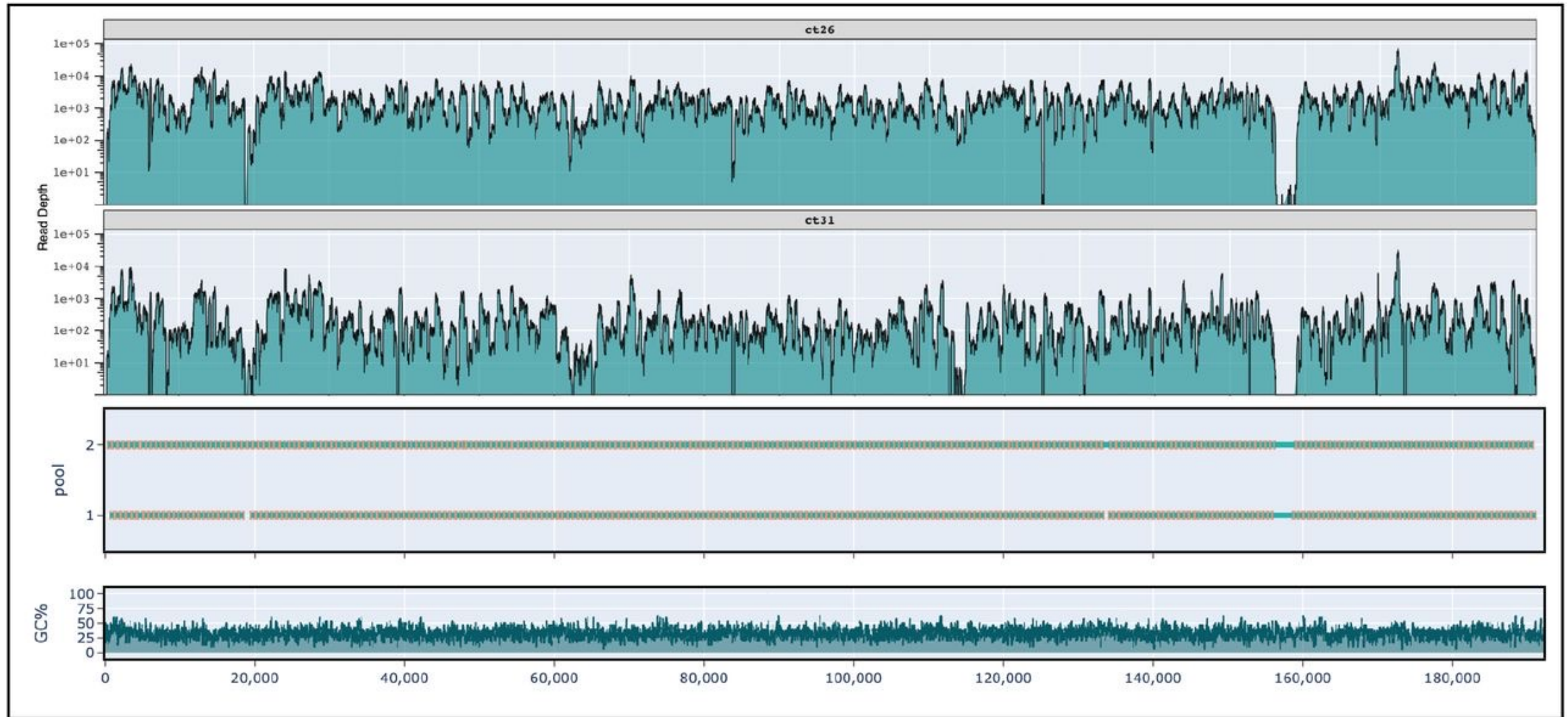


Figure 2: Scheme design and sequencing coverage of MeV primer scheme. The top panel shows the read depth across the MeV genome for each genotype, A in red, B3 in green, D4 in blue, and D5 in purple. The second, third and fourth panel shows the positions of the amplicons, GC content and Shannon entropy across the genome. The fifth panel shows genes (yellow) and the CDS (blue). All features are relative to the coordinates of reference genome NC_001498.



New Results

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PrimalScheme: open-source community resources for low-cost viral genome sequencing

 Chris Kent,  Andrew D Smith,  John Tyson,  Dominika Stepniak,  Eddy Kinganda-Lusamaki,  Tracy Lee,  Mia Weaver,  Natalie Sparks,  Thomas Brier,  Lauren Landsdowne,  Sam Wilkinson,  Rachel Colquhoun,  Áine O'Toole,  Placide Mbala-Kingebeni,  Ian Goodfellow,  Andrew Rambaut,  Nicholas Loman,  Joshua Quick

doi: <https://doi.org/10.1101/2024.12.20.629611>

This article is a preprint and has not been certified by peer review [what does this mean?].

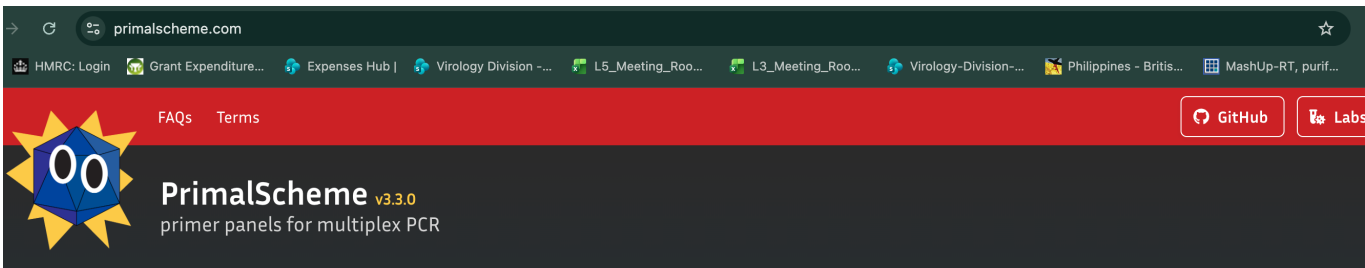
Abstract

Full Text

Info/History

Metrics

 Preview PDF



Chris Kent

Essentials

Scheme name *

Lowercase letters, numbers, and hyphens only. Your input will be automatically formatted to meet these requirements.

Amplicon length *

Min/max length will be ±10%.

Number of pools *

How many pools should the scheme use?

Email *

☐ I understand that any outputs are licensed by CC BY-SA 4.0 *

Proceed to file upload

Fine tuning

Minimum base frequency *

What frequency should a mutation be to be included in primer design?

Target overlap (bp)

Default is 10% amp.

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authors: quick-lab, artic-network

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authors: artic-network

species: 11320

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Acknowledgements

The creation of this training material was commissioned by ECDC to Institut Pasteur with the direct involvement of <name of speaker(s)>