



7- Oxford Nanopore Technologies Additional Details & Troubleshooting

DARRELL L. DINWIDDIE, PHD

DARYL B. DOMMAN, PHD

ONT Flowcell Update

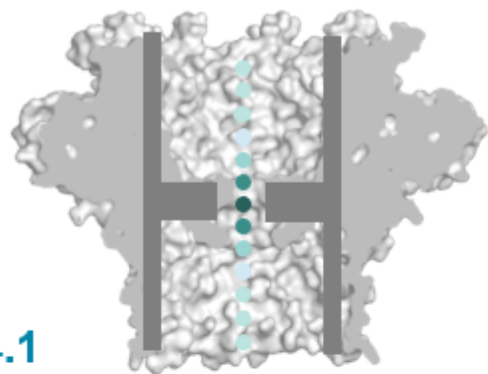


R9.4 vs R10.4 Flowcells

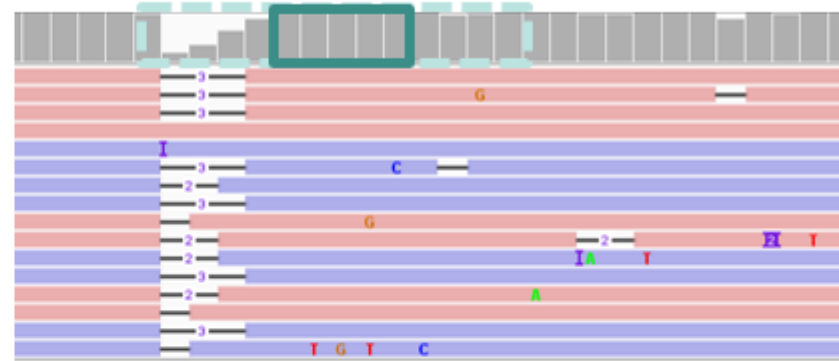
- ▶ R10 is our nanopore chemistry designed to deliver highest consensus accuracy. Paired with the Kit 12 (also referred to as Q20+) chemistry, R10.4 generates data at a modal accuracy above 99%.
- ▶ It contains the proprietary sensor array, Application-Specific Integrated Circuit (ASIC), and R10 nanopores. The R10 series of nanopores contains a double reader-head, and is suitable for experiments where high consensus accuracy is required.
- ▶ **Note: R10.4 flow cells currently require Kit 12 chemistry.**
- ▶ **Currently only available as an Early Access product**

ONT Flowcells

R9.4.1

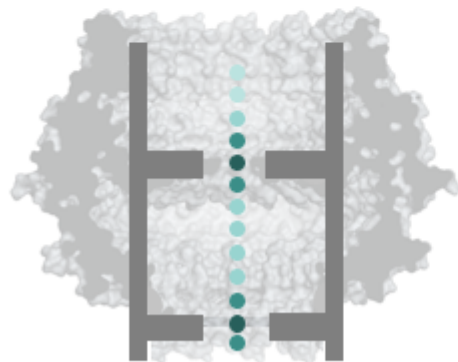


ATCGGAAAAAAAAAATCACGCCACGTCCAAA



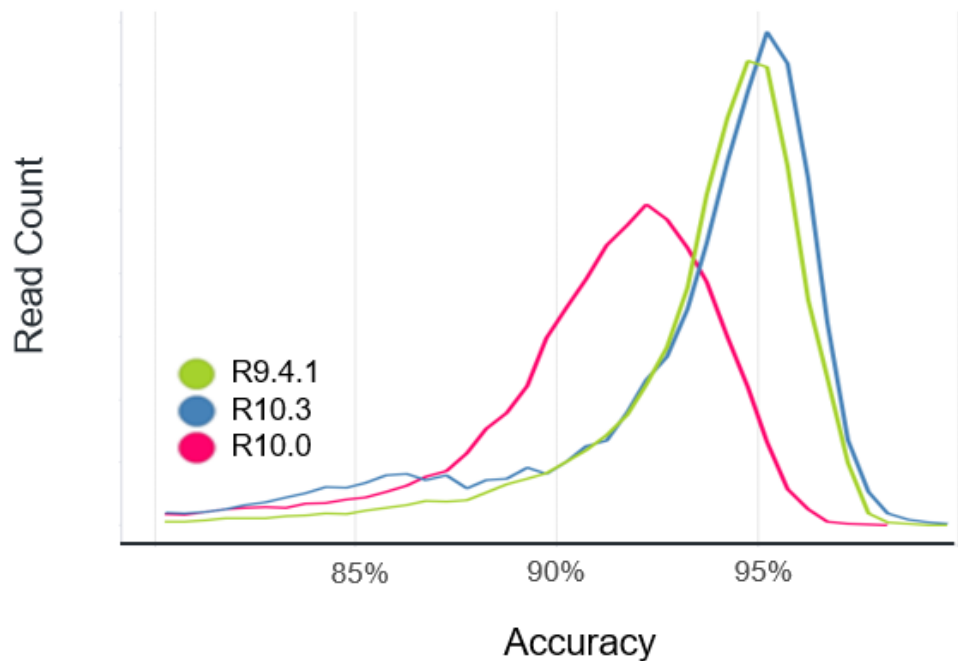
Double
reader-head

R10

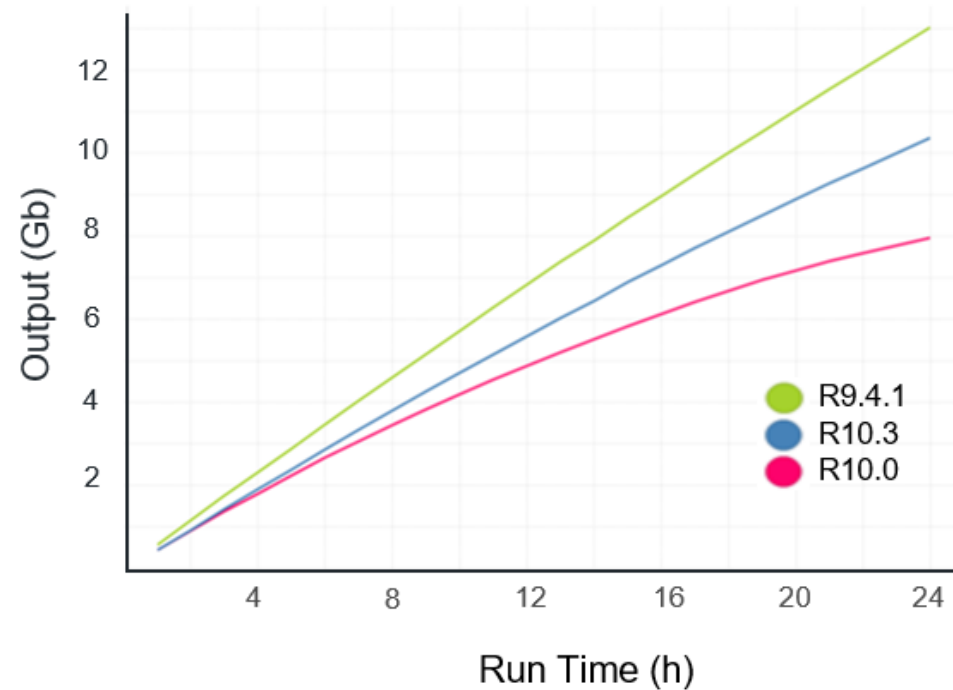


ONT R10 Flowcell

1D Raw read accuracy



MinION data output



Input Requirements:

R9.4.1

R10.3

R10.0

5 – 50 fmol

25 – 75 fmol

50 – 100 fmol

ONT MinKNOW



Component	Recommended specification: GPU high-accuracy basecalling	Minimal specification: data acquisition/CPU basecalling (note: CPU basecalling performance is limited - a GPU is recommended)
Operating system	Windows - 10 Linux - Ubuntu 18.04	Windows - 10 OSX - Mojave, Catalina Linux - Ubuntu 18.04
Memory/RAM	16 GB RAM	16 GB RAM
CPU	Intel i7†, i9, Xeon, or better Ryzen 5, 7, or better, with at least 4 cores/8 threads	Intel i7†, i9, Xeon, or better Ryzen 5, 7, or better, with at least 4 cores/8 threads
GPU	NVIDIA GPU RTX 2060 or better, with at least 8 GB of GPU memory. Theoretical performance is only a guide, but more is better. Technical information can be found on various websites, for example https://www.techpowerup.com/gpu-specs/ . Widely-available examples include the RTX 2060, RTX 2070, RTX 3060, RTX 3070. If you are working with a different type of GPU than the models listed above, please ensure that it has a CUDA Compute Capability >6.1 (for more information about CUDA-enabled GPUs, see the NVIDIA website).	-
Storage	1 TB internal SSD	1 TB internal SSD
Ports	USB3.0‡	USB3.0‡

† Users need to verify that their i7 is a four-core model or better

‡ The MinION device is CE marked using USB3.0. If a user wished to use USB-C, they may, but this invalidates the CE marking

Storage Requirements

Typically, 1 Gbase of sequence data takes up approximately 11 Gbytes of storage. This typically comprises 90% .fast5 files, 9% FASTQ files and 1% sequence summary file.

Example file sizes below are based on different outputs from a flow cell, with a run saving both .fast5 and FASTQ files with a read N50 of 25 kb.

Output (Gbases)	.fast5 storage (Gbytes)	FASTQ storage (Gbytes)	.fast5 + FASTQ storage (Gbytes)
10	100	10	110
15	150	15	165
30	300	30	330

As an experiment progresses, .fast5 files are produced for all reads. If basecalling is chosen, these reads are utilised by the onboard software (more information below) to generate sequence data which is then stored in FASTQ files and as a FASTQ record within the .fast5 file.



MinION



MinION_{Mk1C}

MinKNOW

Log in with your Nanopore account [?](#)

Log in with your Nanopore account

Continue as guest





Start

Sequencing overview

Experiments

System messages

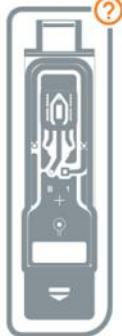
Host settings



Application settings

Connection manager

MN31043 FA-00503

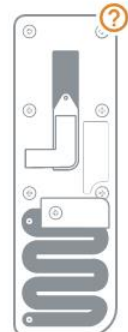


Flow cell not checked

MinION
Detected

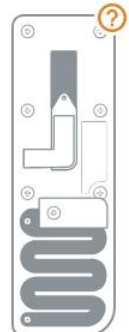
- Start
- Sequencing overview
- Experiments
- System messages
- Host settings

X1 FAH80885




Flow cell not checked

X2 FAP50597



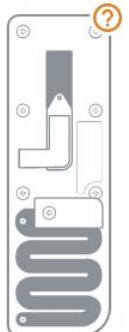
Flow cell not checked

X3



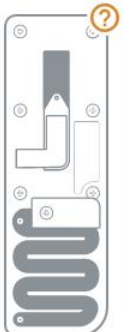
Flow cell not checked

X4 FAL14709



Flow cell not checked

X5 FAO29860



Flow cell not checked

GridION
5 Flowcells



Start Screen

Start sequencing

Analysis

Flow cell check

Hardware check

More

Hardware Check





Hardware check



5 of 5 positions selected

✓ Ready to start

[Select all available](#)



Please ensure that the configuration test cell has been inserted correctly.

Checking
Hardware
Use Test Cell

< Back to start

▶ Start





X1	FAH80885	X2	FAP50597	X3	FAL14709	X5	FAO29860
Checking hardware	Checking hardware	Checking hardware	Checking hardware	Checking hardware	Checking hardware	Checking hardware	Checking hardware

**Checking Hardware
Use Test Cell**




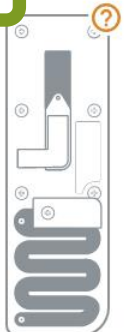


X1  FAH80885




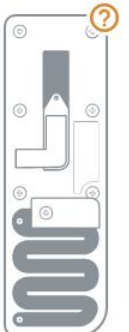
Flow cell not checked

X2  FAP50597




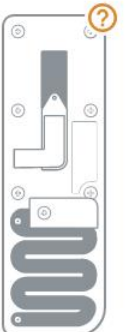
Flow cell not checked

X3  FAH80885




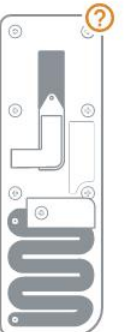
Flow cell not checked

X4  FAL14709



Flow cell not checked

X5  FAO29860



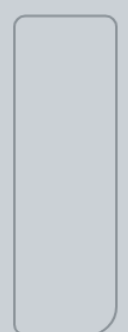
Flow cell not checked

Green check if hardware is working



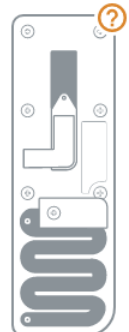
- Start
- Sequencing overview
- Experiments
- System messages
- Host settings

X1



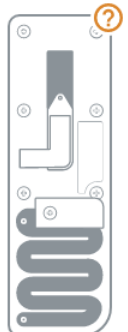
No flow cell detected

X2 FAP50597



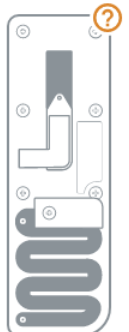
Flow cell not checked

X3



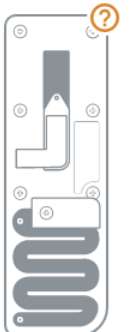
Flow cell not checked

X4 FAL14709



Flow cell not checked

X5 FAO29860



Flow cell not checked

Does not detect
flowcell or test

Restart system



Start Screen

Start sequencing

Analysis

Flow Cell Check



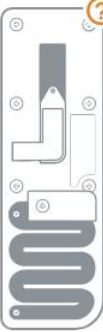



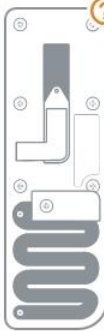
Flow cell check

Hardware check

More



- Start
- Sequencing overview
- Experiments
- System messages
- Host settings

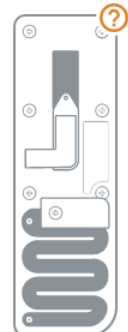
X1	X2	X3	X4	X5
FAH80885	FAR55844	FAL14709	FAL14709	FAO29860
				
Flow cell not checked	Checking flow cell	Flow cell not checked	Flow cell not checked	Flow cell not checked

Flowcell ID

Warms to 37C
Checks for Pores
~20 minutes

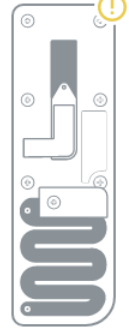
- Start
- Sequencing overview
- Experiments
- System messages
- Host settings

X1 FAH80885



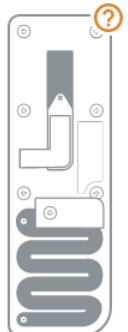
Flow cell not checked

X2 FAR55844



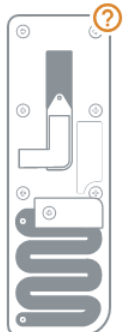
Flow cell below warranty level
564 pores found

X3



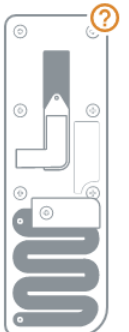
Flow cell not checked

X4 FAL14709



Flow cell not checked

X5 FAO29860



Flow cell not checked

Pores Found

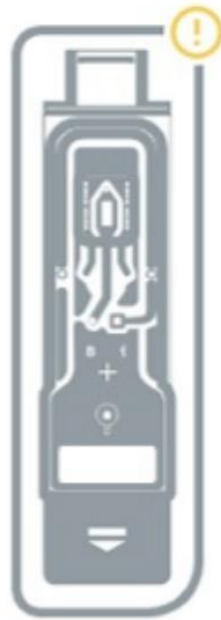
12/4/2022: Flowcell used
12/6/2022: Washed, 948 pores
Stored at 4C
5/16/2022: 564 pores

~60% of pores still active after 6 months of storage

**Failed
Flowcell
Check**

MN31043

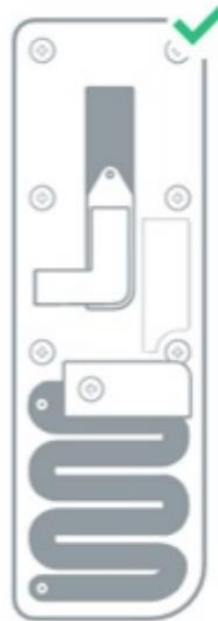
AAA111
FA-00503



Flow cell below warranty level
27 pores found

MN31043

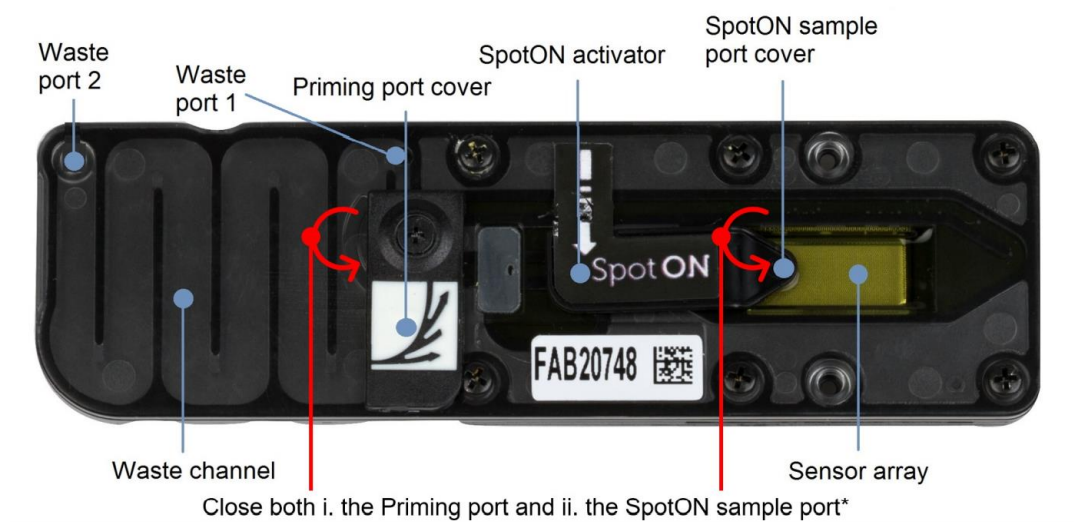
FAO39517



Ready for sequencing
1490 pores found

Flow Cell Pores

Flow cell	Minimum number of active pores covered by warranty
Flongle Flow Cell	50
MinION/GridION Flow Cell	800
PromethION Flow Cell	5000



*Both ports are shown in a closed position



Start Run



Start sequencing

Analysis

Flow cell check

Hardware check

More



- Start
- Sequencing overview
- Experiments
- System messages
- Host settings

1. Positions 3. Run options 4. Basecalling 5. Output 6. Review

Name Run

Select positions

Ensure the experiment name does not contain any personally-identifiable information.

Test-1

Join existing Load settings from template

Select Position

▶

Select all available

Optional Sample ID

Position	Flow cell ID	Flow cell type	Sample ID
X2	FAR55844	FLO-MIN106	Sample ID

Back to start

Continue to kit selection

Skip to final review

GXB03497

My device

Start

1. Positions

2. Kit

3. Run options

4. Basecalling

5. Output

6. Review

Sequencing overview

Experiments

System messages

Host settings

Kit selection

Sample type

DNA

RNA

PCR-free

PCR

PCR-free

Multiplexing

Yes

No

 Control[Reset filters](#)

Select Kit &
Workflow

Ligation Sequencing Kit SQK-LSK109	Rapid Barcoding Kit SQK-RBK004	Rapid Sequencing Kit SQK-RAD004	Direct RNA Sequencing Kit SQK-RNA002
PCR Barcoding Kit SQK-PBK004	16S Barcoding Kit (BC1-24) SQK-16S024	CAS109 Sequencing Protocol SQK-CAS109	CAS109 Sequencing Kit SQK-CS9109
Direct cDNA Sequencing Kit SQK-DCS109	Field Sequencing Kit SQK-LRK001	Ligation Sequencing Kit (48 reactions) SQK-LSK109-XL	Ligation Sequencing Kit SQK-LSK110
Ligation Sequencing Kit XL SQK-LSK110-XL	Ligation Sequencing Kit SQK-LSK112	Ligation Sequencing Kit XL SQK-LSK112-XL	Multiplex Ligation Sequencing Kit XL SQK-MLK110-96-XL
Multiplex Ligation Sequencing Kit XL SQK-MLK111-96-XL	Native Barcoding Sequencing Kit 24 SQK-NBD110-24	Native Barcoding Sequencing Kit 96 SQK-NBD110-96	Native Barcoding Sequencing Kit 24 SQK-NBD112-24
Native Barcoding Sequencing Kit 96 SQK-NBD112-96	PCR cDNA Barcoding Kits SQK-PCB109	PCR-cDNA Barcoding Kit 24 SQK-PCB110-24	PCR cDNA Barcoding Kit - 24 SQK-PCB111-24
PCR-cDNA Sequencing Kit SQK-PCS109	PCR cDNA Sequencing Kit SQK-PCS110	PCR cDNA Sequencing Kit SQK-PCS111	Pore-C Sequencing Protocol SQK-PRC109
PCR Sequencing Kit SQK-PSK004	Q20 Early Access Kit SQK-Q20EA	Rapid Sequencing Kit SQK-RAD112	Rapid Barcoding Kit 96 SQK-RBK110-96
Rapid PCR Barcoding Kit SQK-RPB004	Ultra-Long DNA Sequencing Kit SQK-ULK001	VoITRAX PCR Tiling 1-12 COVID-19 VSK-PTC001	VoITRAX Multiplex Sequencing Kit VSK-VMK002
VoITRAX Multiplex Sequencing Kit VSK-VMK003	VoITRAX Sequencing Kit VSK-VSK002	VoITRAX Sequencing Kit VSK-VSK003	16S Barcoding Kit SQK-RAB204

[Back to position selection](#)[Continue to run options](#)[Skip to final review](#)

Connection manager

- Start
- Sequencing overview
- Experiments
- System messages
- Host settings

Kit selection

Sample type:

PCR-free:

Multiplexing: Control

[Reset filters](#)

Ligation Sequencing Kit SQK-LSK109	Rapid Barcoding Kit SQK-RBK004	Rapid Sequencing Kit SQK-RAD004	Direct RNA Sequencing Kit SQK-RNA002
PCR Barcoding Kit SQK-PBK004	16S Barcoding Kit (BC1-24) SQK-16S024	CAS109 Sequencing Protocol SQK-CAS109	CAS109 Sequencing Kit SQK-CS9109
Direct cDNA Sequencing Kit SQK-DCS109	Field Sequencing Kit SQK-LRK001	Ligation Sequencing Kit (48 reactions) SQK-LSK109-XL	Ligation Sequencing Kit SQK-LSK110
Ligation Sequencing Kit XL SQK-LSK110-XL	Ligation Sequencing Kit SQK-LSK112	Ligation Sequencing Kit XL SQK-LSK112-XL	Multiplex Ligation Sequencing Kit XL SQK-MLK110-96-XL
Multiplex Ligation Sequencing Kit XL SQK-MLK111-96-XL	Native Barcoding Sequencing Kit 24 SQK-NBD110-24	Native Barcoding Sequencing Kit 96 SQK-NBD110-96	Native Barcoding Sequencing Kit 24 SQK-NBD112-24
Native Barcoding Sequencing Kit 96 SQK-NBD112-96	PCR cDNA Barcoding Kits SQK-PCB109	PCR-cDNA Barcoding Kit 24 SQK-PCB110-24	PCR cDNA Barcoding Kit - 24 SQK-PCB111-24
PCR-cDNA Sequencing Kit SQK-PCS109	PCR cDNA Sequencing Kit SQK-PCS110	PCR cDNA Sequencing Kit SQK-PCS111	Pore-C Sequencing Protocol SQK-PRC109
PCR Sequencing Kit SQK-PSK004	Q20 Early Access Kit SQK-Q20EA	Rapid Sequencing Kit SQK-RAD112	Rapid Barcoding Kit 96 SQK-RBK110-96 ✓
Rapid PCR Barcoding Kit SQK-RPB004	Ultra-Long DNA Sequencing Kit SQK-ULK001	VolTRAX PCR Tiling 1-12 COVID-19 YSK-PTC001	VolTRAX Multiplex Sequencing Kit YSK-VMK002

Select expansion pack

Midnight RT PCR Expansion
EXP-MRT001

Spike Seq RT PCR Expansion
EXP-SRT001

**SARS-CoV-2
Midnight Primers
Rapid Barcoding**

← Back to position selection

Continue to run options >

Skip to final review >>

- Start
- Sequencing overview
- Experiments
- System messages
- Host settings

Run options

Run length ?

Bias voltage ?

- 72 hours +

- -180 mV +

Adaptive sampling ?

Disabled Options

> Show advanced user options

**Adjust Run Time
Normally 24-36 hrs
max**

- Start
- Sequencing overview
- Experiments
- System messages
- Host settings

Basecalling

Basecalling ?

Config: High-accuracy basecalling Options

Barcoding ?

Enabled Options

Alignment ?

Disabled Options

Only use high accuracy basecalling if running on GPU

Poor Performance Using High Accuracy Basecalling on Mk1C

- Start
- Sequencing overview
- Experiments
- System messages
- Host settings

- 1. Positions
- 2. Kit
- 3. Run options
- 4. Basecalling
- 5. Output
- 6. Review

Output

Output location ⓘ

/data/

Output format ⓘ

FAST5 FASTQ BAM

Filtering ⓘ

Qscore: 9 | Readlength: Unfiltered | Read splitting: Disabled Options

> Show advanced user options

Normally run with default (both fast5 & fastq) selected

- Start
- Sequencing overview
- Experiments
- System messages
- Host settings

- 1. Positions
- 2. Kit
- 3. Run options
- 4. Basecalling
- 5. Output
- 6. Review

Normally run with default (both fast5 & fastq) selected

Test-1

Selected positions

Kit

Selected kit: SQK-RBK110-96
Expansion packs: EXP-MRT001

Edit

Run options

Run length: 72 hours
Bias voltage: -180 mV
Adaptive sampling: Off

Advanced run options

Basecalling

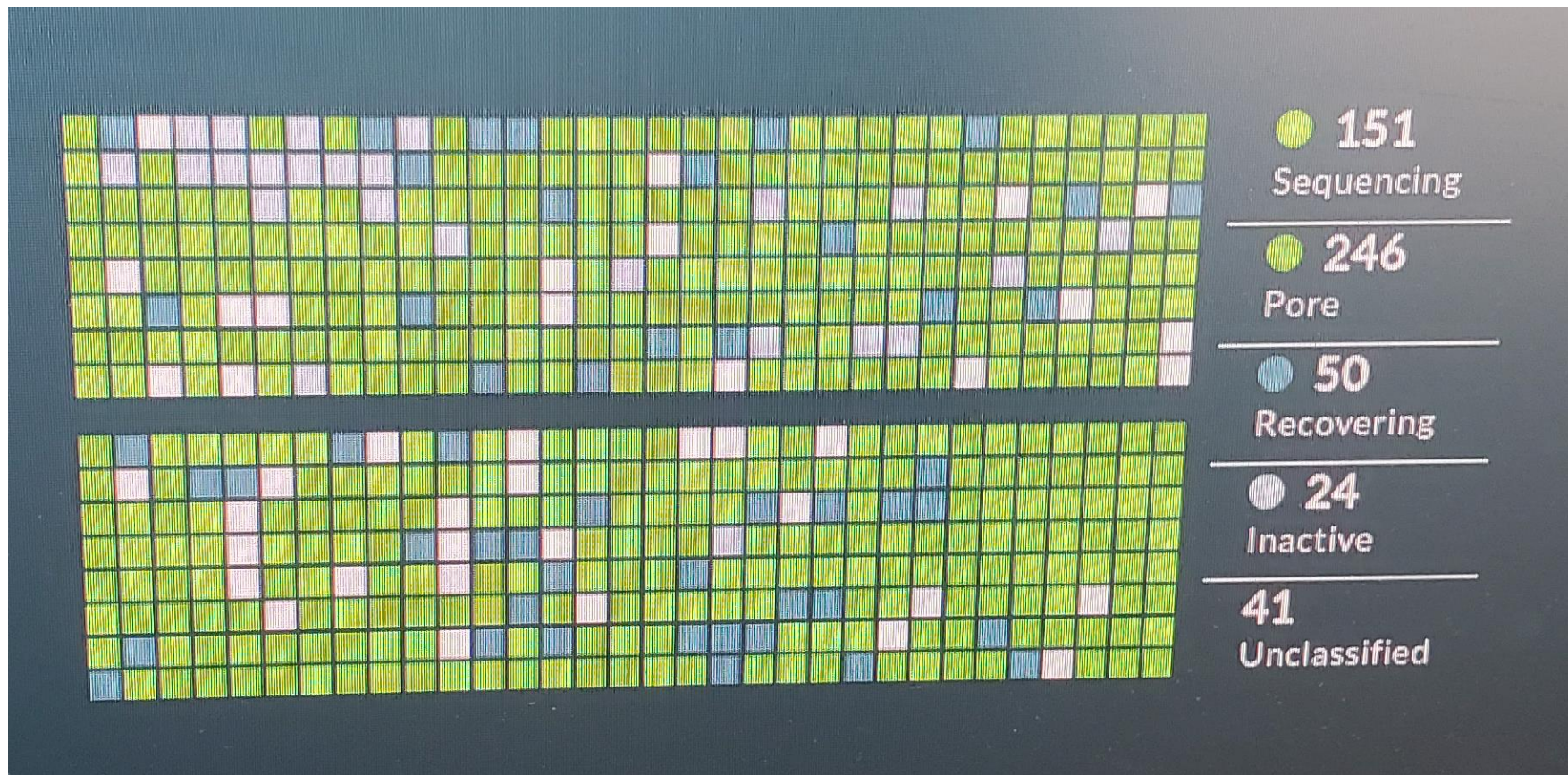
Basecalling: On (High-accuracy basecalling)
Barcoding: On
Alignment: Off

Output

Location: /data/
FAST5: On (Raw, FASTQ record, VBZ, 4000 reads per ...
FASTQ: On (GZip, 4000 reads per file)
Read filtering: Qscore: 9 | Readlength: Unfiltered | Read split...

Advanced run options

Pore Channels Panel

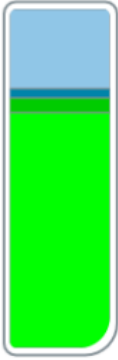


GXB03153 REMOTE
gxb03153

Guest

- Start
- Sequencing overview
- Experiments
- System messages
- Host settings >


X1 FAQ16580



Sequencing

Detailed description: A vertical rectangular flow cell icon with a blue top section and a green bottom section, indicating it is in use for sequencing.


X2 FAQ13638



Sequencing

Detailed description: A vertical rectangular flow cell icon with a blue top section and a green bottom section, indicating it is in use for sequencing.


X3



No flow cell detected

Detailed description: A vertical rectangular flow cell icon that is completely greyed out, indicating no flow cell is present.


X4



No flow cell detected

Detailed description: A vertical rectangular flow cell icon that is completely greyed out, indicating no flow cell is present.

X5



No flow cell detected

Detailed description: A vertical rectangular flow cell icon that is completely greyed out, indicating no flow cell is present.

Overview of
flowcells
sequencing

GXB02294 REMOTE

GXB02294

SD

Start

Sequencing overview

Experiments

System messages

Host settings

Experiments last 7 days

Search experiments

RR_Ladder

OP_TEST

Workflow 1

RR_Midnight_22_03

Resume

Pause

Stop

Start pore scan

Export run report

Experiment group

Position	Flow cell ID	Sample ID	Health	Run time	Run state	Reads	Bases	Basecalled %
X4	FAO29273	no_sample		15 h / 72 h	Active	5.32 M	1.3 Gb basecalled 1.46 Gb estimated	100%
X5	FAS02937	no_sample		15 h / 72 h	Active	4.27 M	1.05 Gb basecalled 1.17 Gb estimated	100%

Overview of
flowcells
sequencing

Note: Poor
sequencing
on X4

- Start
- Sequencing overview
- Experiments
- System messages
- Host settings

Filters

4_3_basecalling_test

Reads: 236.22 k

Estimated bases: 1.07 Gb

Basecalled bases: 1.11 Gb

Active runs: 2

Total runs: 4

▶ Resume
⏸ Pause
■ Stop
~ Start pore scan
↓ Export run report
~ Experiment group

Position	Flow cell ID	Sample ID	Health	Run time	Run state	Reads	Bases	Basecalled %
X1	FAQ16580	fast_basecalling	<div style="width: 80%; background-color: #28a745; height: 10px;"></div>	16 m / 72 h	Active	19.65 k	86.77 Mb basecalled 84.64 Mb estimated	100%
X2	FAQ13638	fast_basecalling	<div style="width: 80%; background-color: #28a745; height: 10px;"></div>	16 m / 72 h	Active	20.96 k	97.4 Mb basecalled 94.12 Mb estimated	100%
X1	FAQ16580	no_sample	—	1 h 1 m	Complete	89.91 k	413.74 Mb basecalled 399.62 Mb estimated	100%
X2	FAQ13638	no_sample	—	1 h 1 m	Complete	105.7 k	510.42 Mb basecalled 487.38 Mb estimated	100%

< Back to main menu

☰ Device settings

📁 Software

📁 File manager

⚙️ System

❓ Help

Device settings

System

GXB03214

🔌 Shut down

Date and time

Wed 2022-03-23 13:27:01 Europe/London (time.oxfordnanolabs.local)

📅 Change date and time

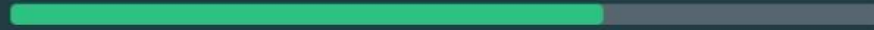
🌐 Change time zone

Disk management

➕ Add network drive

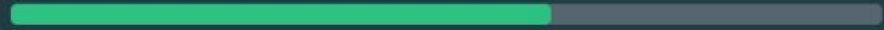
Internal drive

System storage



/data

🔗 Share



Check for hard
drive space
before starting run

GXB03214 REMOTE

Host settings

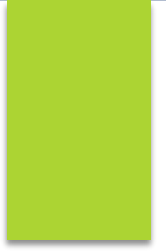
- Back to main menu
- Device settings
- Software**
- File manager
- System
- Help

Software

MinKNOW	Installed version: 22.a.33 (xenial) Available version: 22.a.35 (xenial)	Install update
System	Total packages: 121 Security packages: 75 <small>If you see some system packages remain after a system update, please update MinKNOW and the number should return to zero.</small>	Install packages

Check for software updates before starting run

ONT Data Output



- Recent
- Home
- Desktop
- Documents
- Downloads
- Music
- Pictures
- Videos
- Trash
- Network
- Computer**
- USB-STICK
- Connect to Server

bin 159 items	boot 14 items	data 34 items	dev 210 items
etc 262 items	home 1 item	lib 23 items	lib32 43 items
lib64 1 item	media 9 items	mnt 0 items	opt 4 items
proc 433 items	required-debs 0 items	root	run 56 items
sbin 209 items	snap 1 item	srv 0 items	sys 11 items
tmp 12 items	usr 11 items	var 14 items	initrd.img 64.5 MB



"data" selected (containing 34 items)

- My device
- Start
- Sequencing overview
- Experiments
- System messages
- Host settings

NMCurative289_384

Resume Pause Stop Start MUX scan Export PDF Experiment group

Position	Flow cell ID	Sample ID	Health	Run time	Run state	Reads	Bases	Basecalled %
X2	FAS69901	NMCurative289_384	—	1 d 6 h	Complete	8.39 M	5.34 Gb basecalled 5.49 Gb estimated	100%

report_FAS76468_20220427_1401_f062b3b3.pdf

1 of 13 87.82%

Thumbnail:

GXB03497 (localhost)
X2
TRL1F193_200_3F1_88
TRL1F193_200_3F1_88
f062b3b3-fa62-4dc3-927c-2b979cc3d513

20220421_1041_X2_FAS69938_0c6a18ad

data TRL_1F_001_096 TRL_1F_001_096 20220421_1041...9938_0c6a18ad

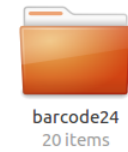
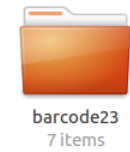
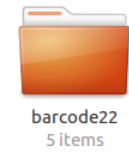
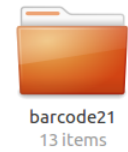
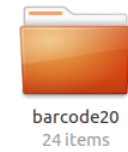
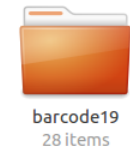
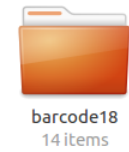
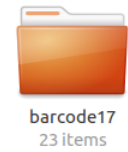
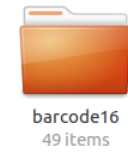
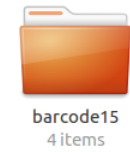
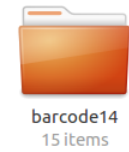
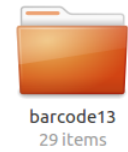
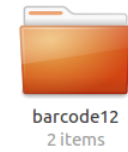
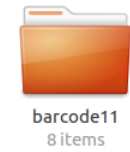
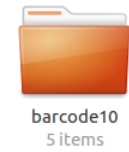
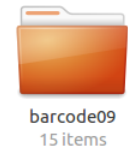
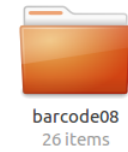
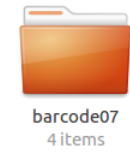
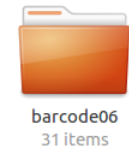
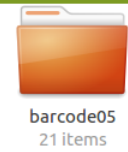
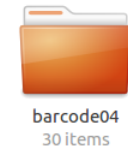
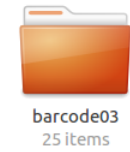
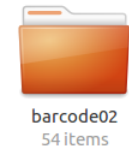
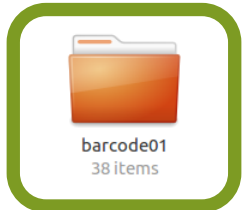
- Recent
- Home
- Desktop
- Documents
- Downloads
- Music
- Pictures
- Videos
- Trash
- Network
- Computer
- Seagate Portable Drive
- USB-STICK
- Connect to Server

fast5_fail fast5_pass fastq_fail **fastq_pass** other_reports barcode_alignment_FAS69938_294a03... duty_time_FAS69938_294a03dc.csv final_summary_FAS69938_294a03dc.txt

report_FAS69938_20220421_1046_0c6a18ad.md report_FAS69938_20220421_1046_0c6a18ad.pdf sequencing_summary_FAS69938_294a03dc.txt throughput_FAS69938_294a03dc.txt

"report_FAS69938_20220421_1046_0c6a18ad.pdf" selected (567.2 kB)

- Recent
- Home
- Desktop
- Documents
- Downloads
- Music
- Pictures
- Videos
- Trash
- Network
- Computer
- USB-STICK
- Connect to Server



- Recent
- Home
- Desktop
- Documents
- Downloads
- Music
- Pictures
- Videos
- Trash
- Network
- Computer
- USB-STICK
- Connect to Server



FAS76468_pass_barcode01_4db4895e_3.8 MB



FAS76468_pass_barcode01_4db4895e_1.fastq.gz 3.8 MB



FAS76468_pass_barcode01_4db4895e_2.fastq.gz 3.9 MB



FAS76468_pass_barcode01_4db4895e_3.fastq.gz 3.9 MB



FAS76468_pass_barcode01_4db4895e_4.fastq.gz 3.9 MB



FAS76468_pass_barcode01_4db4895e_5.fastq.gz 3.9 MB



FAS76468_pass_barcode01_4db4895e_6.fastq.gz 3.9 MB



FAS76468_pass_barcode01_4db4895e_7.fastq.gz 3.9 MB



FAS76468_pass_barcode01_4db4895e_8.fastq.gz 3.9 MB



FAS76468_pass_barcode01_4db4895e_9.fastq.gz 3.8 MB



FAS76468_pass_barcode01_4db4895e_10.fastq.gz 3.9 MB



FAS76468_pass_barcode01_4db4895e_11.fastq.gz 3.9 MB



FAS76468_pass_barcode01_4db4895e_12.fastq.gz 3.9 MB



FAS76468_pass_barcode01_4db4895e_13.fastq.gz 3.9 MB



FAS76468_pass_barcode01_4db4895e_14.fastq.gz 3.9 MB



FAS76468_pass_barcode01_4db4895e_15.fastq.gz 3.9 MB



FAS76468_pass_barcode01_4db4895e_16.fastq.gz 3.9 MB



FAS76468_pass_barcode01_4db4895e_17.fastq.gz 3.9 MB



FAS76468_pass_barcode01_4db4895e_18.fastq.gz 3.8 MB



FAS76468_pass_barcode01_4db4895e_19.fastq.gz 3.9 MB

- My device
- Start
- Sequencing overview
- Experiments
- System messages
- Host settings

NMCurative289_384

Resume
Pause
Stop
Start MUX scan
Export PDF
Experiment group

Position	Flow cell ID	Sample ID	Health	Run time	Run state	Reads	Bases	Basecalled %
X2	FAS69901	NMCurative289_384	—	1 d 6 h	Complete	8.39 M	5.34 Gb basecalled 5.49 Gb estimated	100%

report_FAS76468_20220427_1401_f062b3b3.pdf

1 of 13 87.82%

Thumbnail:

GXB03497 (localhost)
X2
TRL1F193_200_3F1_88
TRL1F193_200_3F1_88
f062b3b3-fa62-4dc3-927c-2b979cc3d513

20220421_1041_X2_FAS69938_0c6a18ad

data TRL_1F_001_096 TRL_1F_001_096 20220421_1041...9938_0c6a18ad

- Recent
- Home
- Desktop
- Documents
- Downloads
- Music
- Pictures
- Videos
- Trash
- Network
- Computer
- Seagate Portable Drive
- USB-STICK
- Connect to Server

fast5_fail fast5_pass fastq_fail fastq_pass other_reports barcode_alignment_FAS69938_294a03... duty_time_FAS69938_294a03dc.csv final_summary_FAS69938_294a03dc.txt

report_FAS69938_20220421_1046_0c6a18ad.md **report_FAS69938_20220421_1046_0c6a18ad.pdf** sequencing_summary_FAS69938_294a03dc.txt throughput_FAS69938_294a03dc.csv

"report_FAS69938_20220421_1046_0c6a18ad.pdf" selected (567.2 kB)

Run Info

Host Name	GXB03497 (localhost)
Position	X3
Experiment Name	TRL3F89_184
Sample ID	TRL3F89_184
Run ID	9c1a2212-3f3d-428b-825f-391a2b6e6492
Acquisition ID(s)	4231a08ef8776a4c19649916575b02b94bd339f2, d67886505f5121d758cdfa3eed9385fb0c40c34
Flow Cell Id	FAS76433
Start Time	April 27, 13:58
Run Length	1d 6h 5m

Run Summary

Reads Generated	12.2 M
Passed Bases	5.77 Gb
Failed Bases	1.59 Gb
Estimated Bases	7.76 Gb

Summary of Run

Run Parameters

Flow Cell Type

FLO-MIN106

Kit

SQK-RBK110-96

Initial bias voltage

-180 mV

FAST5 output

Enabled

FASTQ output

Enabled

BAM output

Disabled

Bulk file output

Disabled

Active channel selection

Enabled

Basecalling

Enabled

Specified run length

30 hours

FAST5 reads per file

4000

FAST5 output options

vbz_compress,fastq,raw

FASTQ reads per file

4000

FASTQ output options

compress

Mux scan period

1 hour 30 minutes

Reserved pores

0 %

Basecall model

High-accuracy basecalling

Barcoding

barcoding_kits=["SQK-RBK110-96"],trim_barcodes="off",require_barcodes_both_ends="off",detect_mid_strand_barcodes="off"

Read filtering

min_qscore=9

Read splitting

enable=off

**Summary of Run
Parameters**

Program Versions

Check for latest software

Versions

MinKNOW

21.11.7

MinKNOW Core

4.5.4

Bream

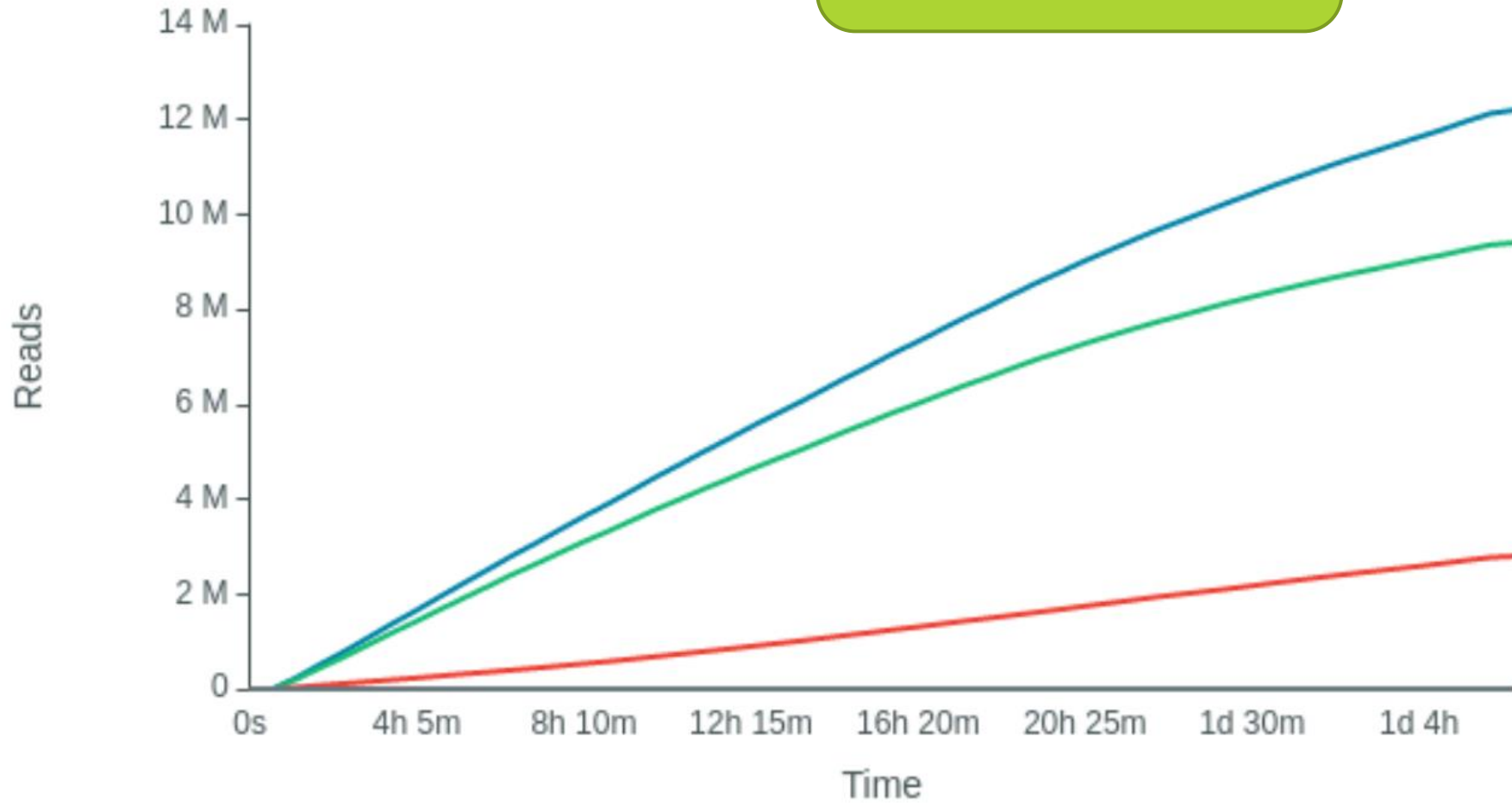
6.3.5

Guppy

5.1.13

Cumulative Output Reads

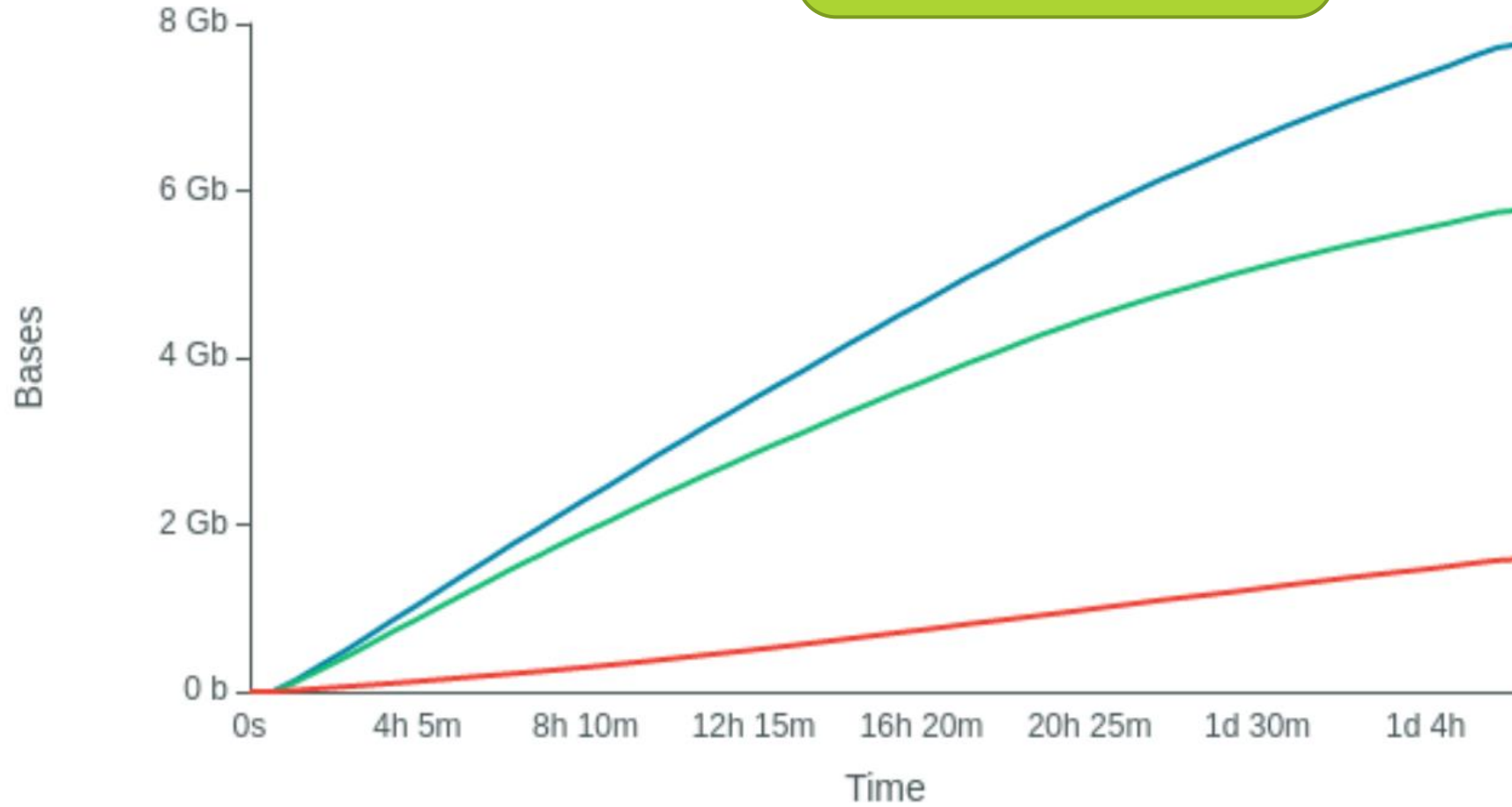
Passing Reads
Flattening After 28 hrs



● Total reads ● Passed reads ● Failed reads ● Skipped reads

Cumulative Output Bases

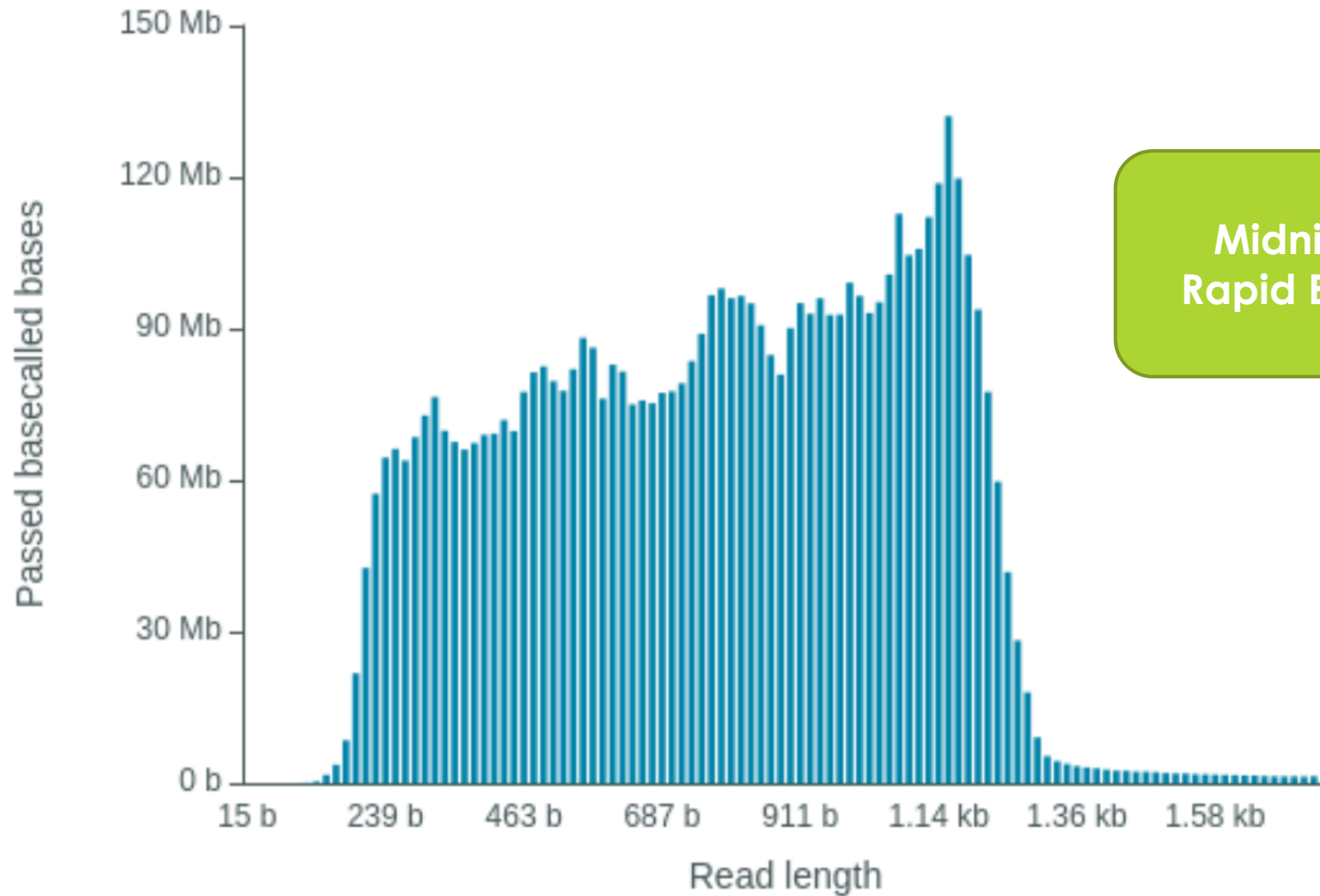
Base Output matches
Read Output



● Estimated bases ● Called bases (passed) ● Called bases (failed)

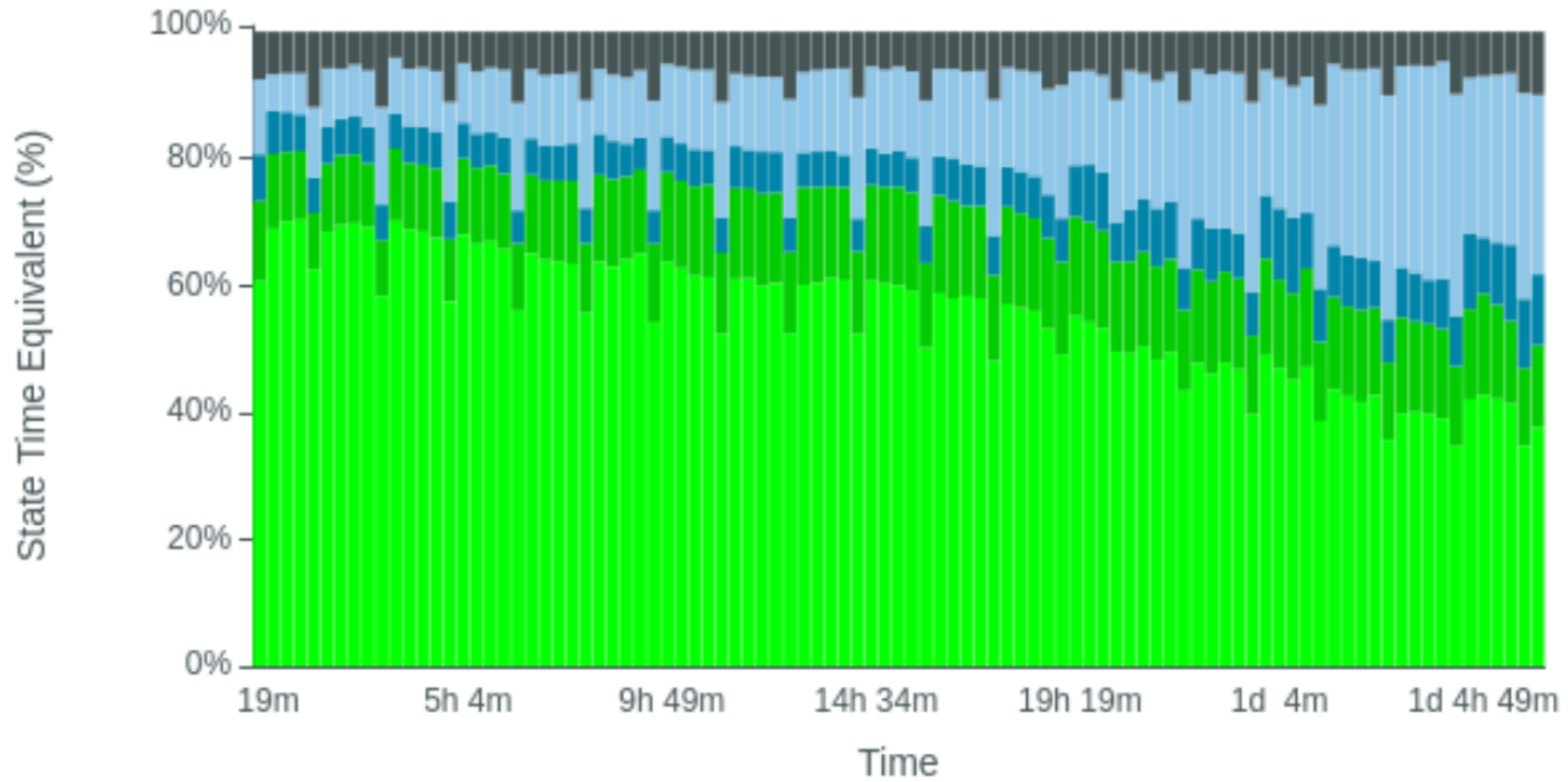
Read Length Histogram Basecalled Bases - Outliers Discarded

Estimated N50: 789 b



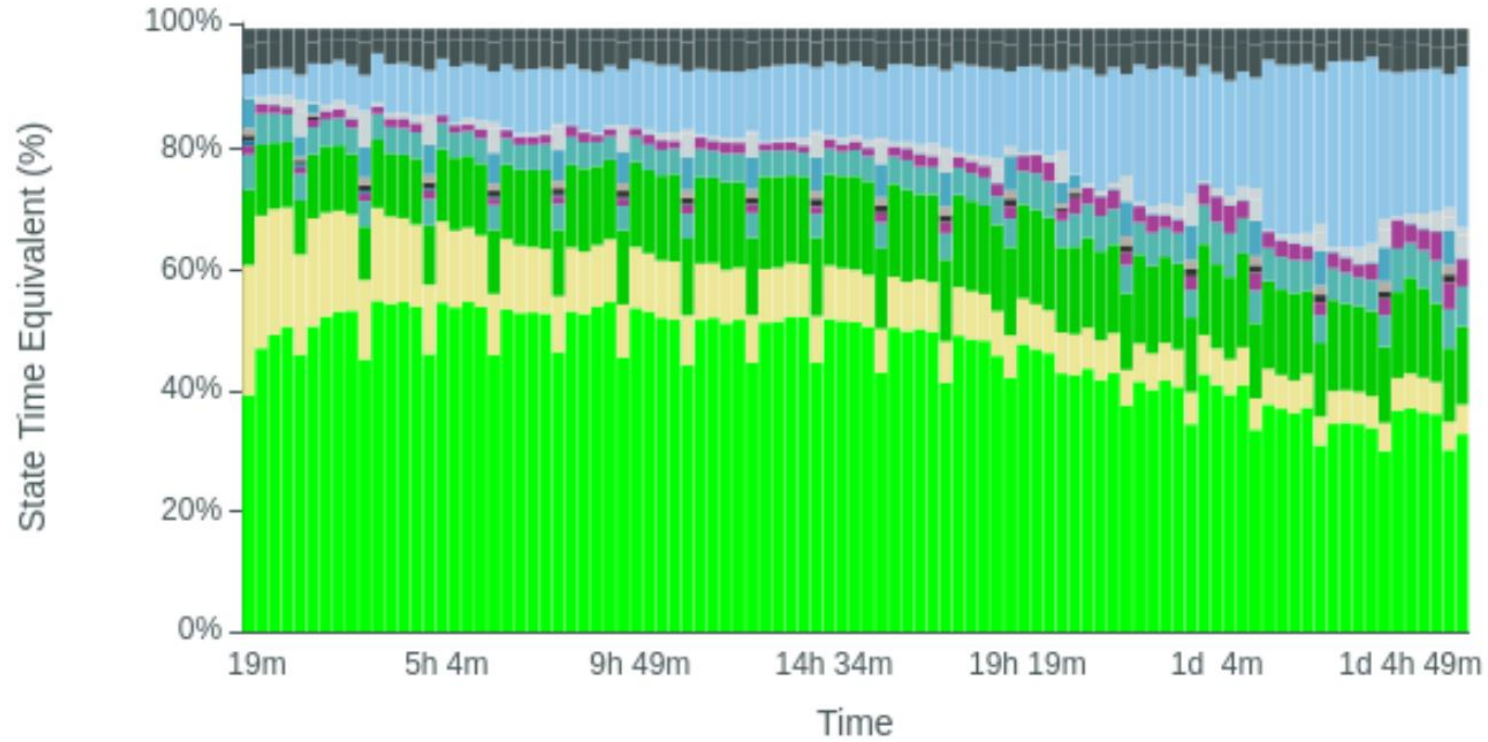
Midnight Primers
Rapid Barcoding Kit

Duty Time Grouped



● Sequencing | ● Pore | ● Recovering | ● Inactive | ● Unclassified

Duty time Categorised



● Strand
● Adapter

● Single Pore

● Unavailable
● Active Feedback

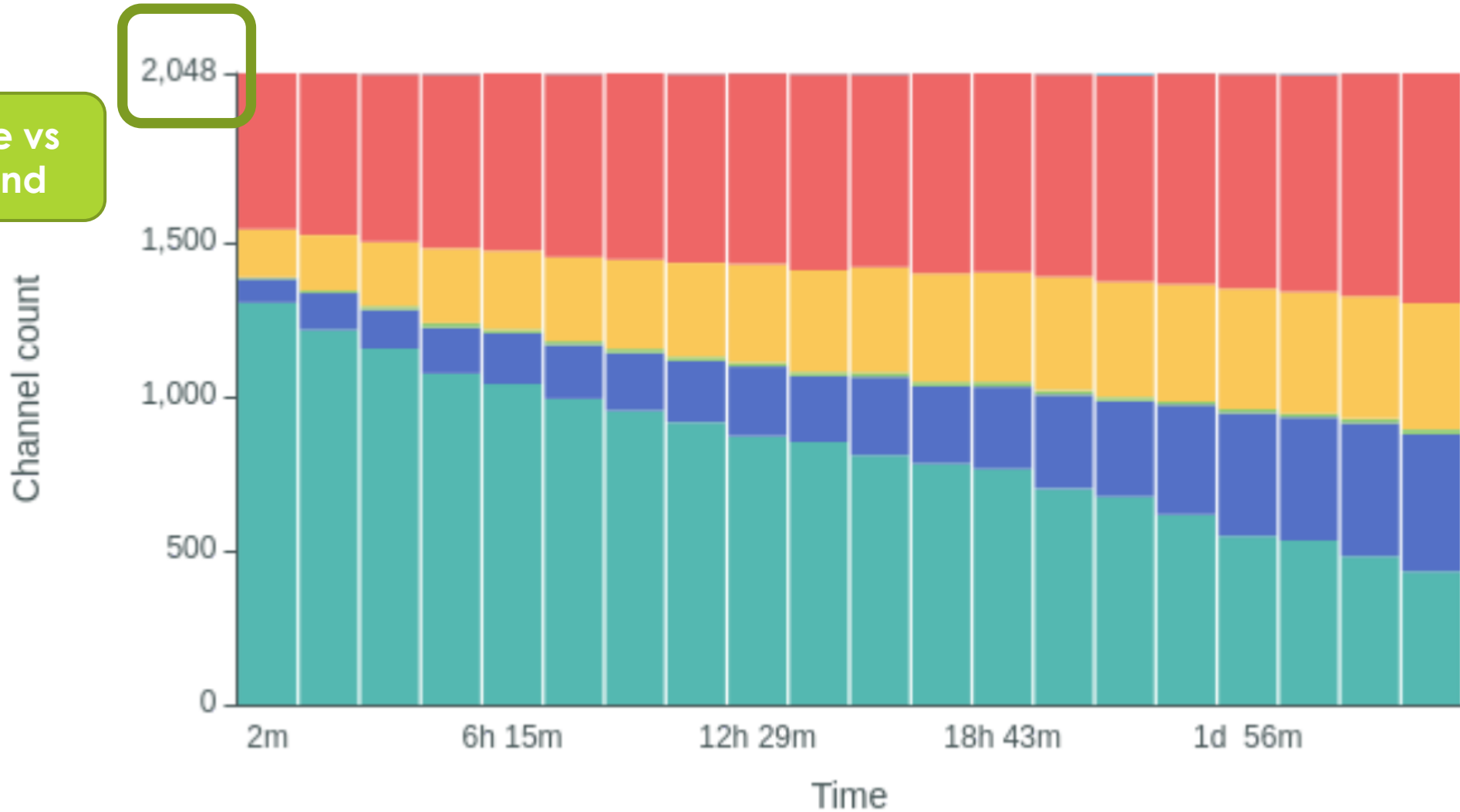
● No Pore From Scan
● Out Of Range 2
● Possible Multiple
● Saturated
● Out Of Range 1
● Zero
● Channel Disabled

● Unclassified
● Unclassified_following_reset
● Pending_manual_reset
● Pending_mux_change
● Pending Reselection

Mux Scan Categorised

● Single Pore ● Reserved Pore ● Unavailable ● Multiple ● Saturated ● Zero ● Other

Note Scale vs Pores Found



Run Debug Messages

- The sequencing run has finished, but basecalling may continue April 28, 20:04
- Mux scan for flow cell FAS76433 has found a total of 681 pores. 375 pores available for immediate sequencing April 28, 19:40
- Performing Mux Scan April 28, 19:38
- Mux scan for flow cell FAS76433 has found a total of 721 pores. 390 pores available for immediate sequencing April 28, 18:07
- Performing Mux Scan April 28, 18:05
- Mux scan for flow cell FAS76433 has found a total of 717 pores. 354 pores available for immediate sequencing April 28, 16:34
- Performing Mux Scan April 28, 16:31
- Mux scan for flow cell FAS76433 has found a total of 748 pores. 368 pores available for immediate sequencing April 28, 15:00

- Performing Mux Scan April 27, 18:43
- Mux scan for flow cell FAS76433 has found a total of 1088 pores. 470 pores available for immediate sequencing April 27, 17:12
- Performing Mux Scan April 27, 17:10
- Mux scan for flow cell FAS76433 has found a total of 1114 pores. 478 pores available for immediate sequencing April 27, 15:38
- Performing Mux Scan April 27, 15:36
- Mux scan for flow cell FAS76433 has found a total of 1199 pores 489 pores available for immediate sequencing April 27, 14:05
- Performing Mux Scan April 27, 14:03
- Starting sequencing procedure April 27, 14:03
- Waiting up to 300 seconds for temperature to stabilise at 34.0°C April 27, 13:58

Pores Found

Questions?

