THE UTILITY OF SARS-COV-2 GENOME SEQUENCING ON THE ION TORRENT GENEXUS PLATFORM

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Division of Laboratory Medicine
10/07/2021

THE UNIVERSITY OF ALABAMA AT BIRMINGHAM

DISCLOSURES

- Principal Investigator on research studies with:
  - Amplyx Pharmaceuticals
  - GenMark Diagnostics
  - CNINE
  - IMMY
  - mFluidX
  - Abnova
  - SpeeDx

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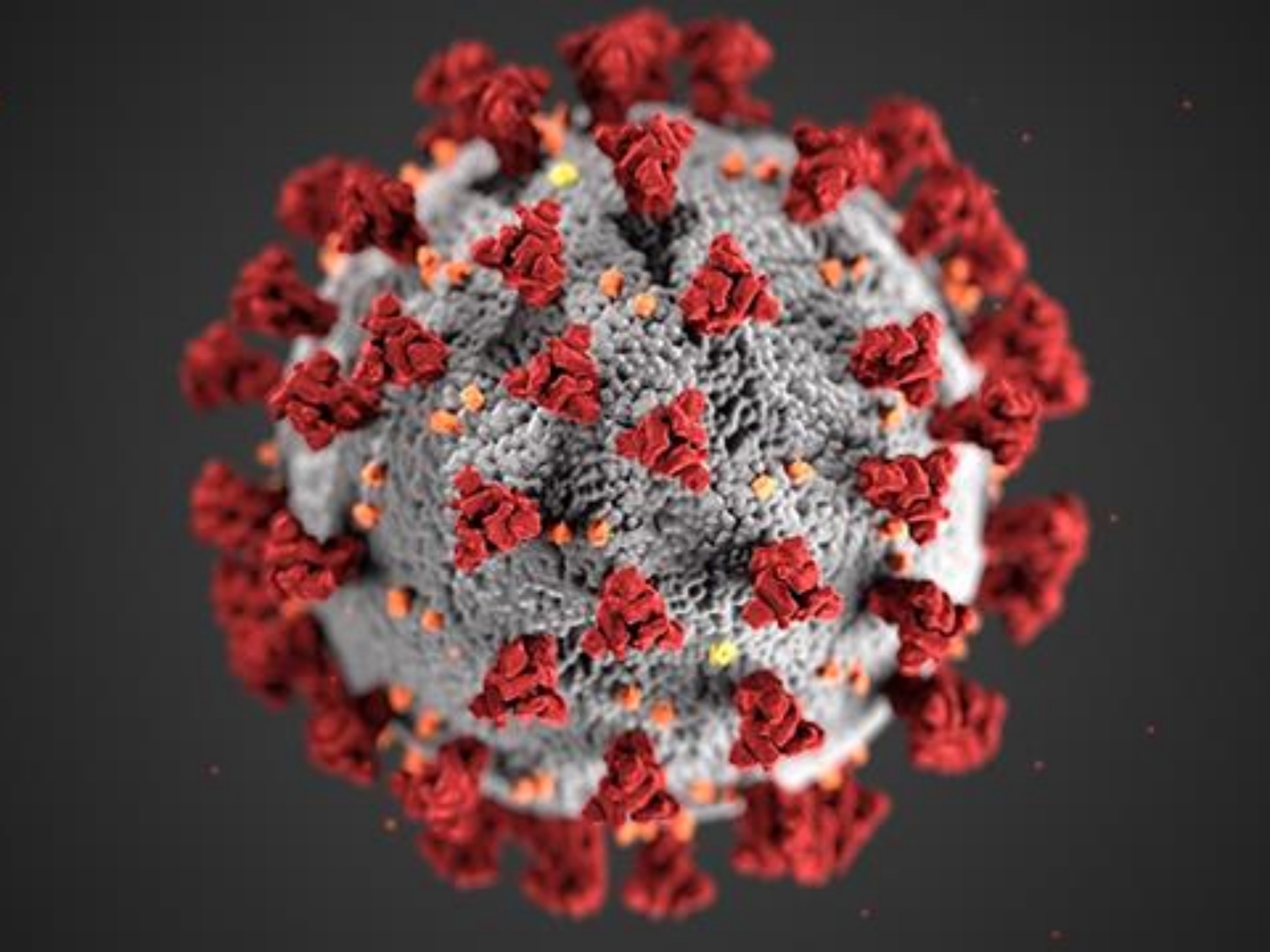
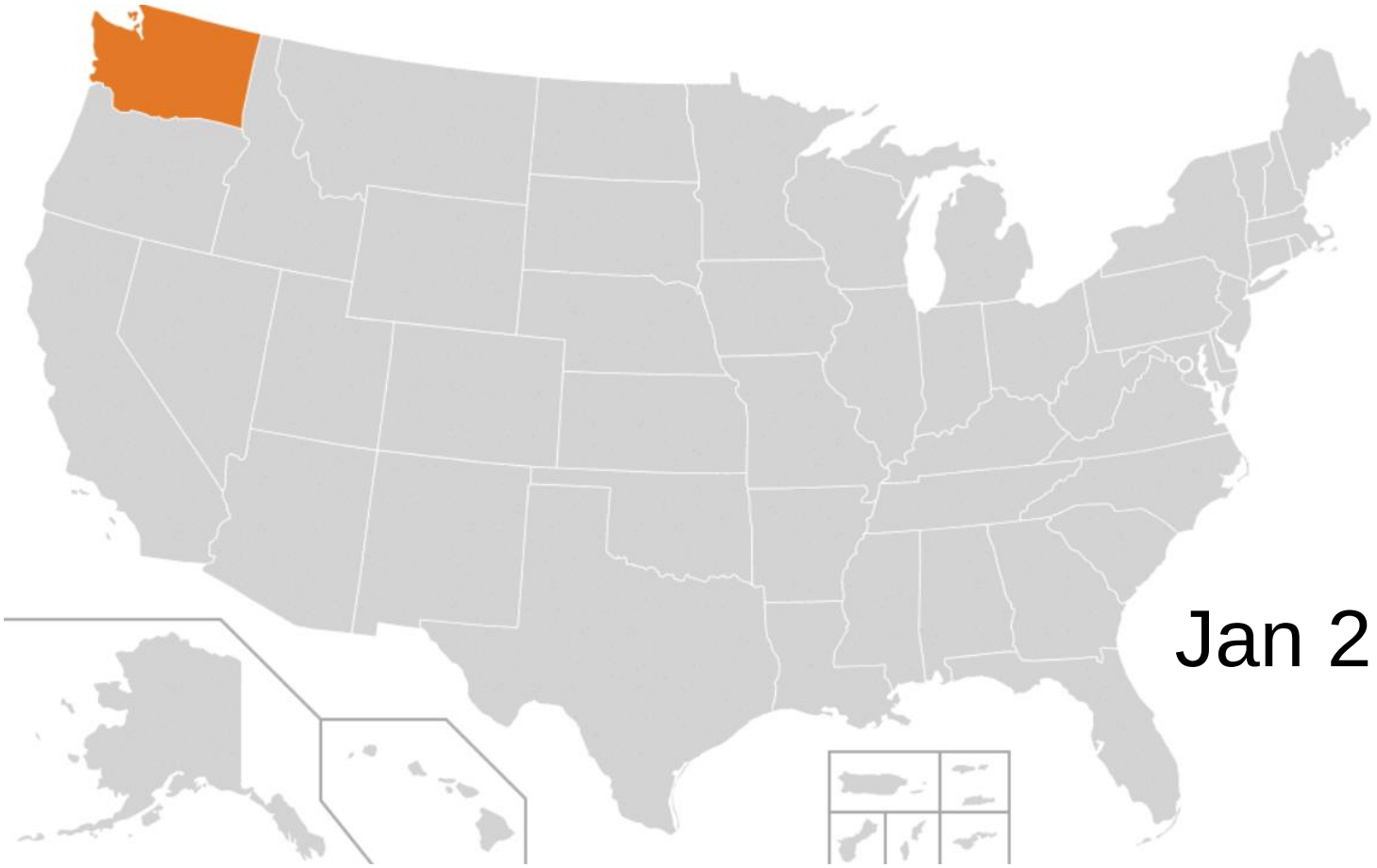
- Speaker was provided honorarium by Thermo Fisher Scientific for providing this presentation.

OBJECTIVES

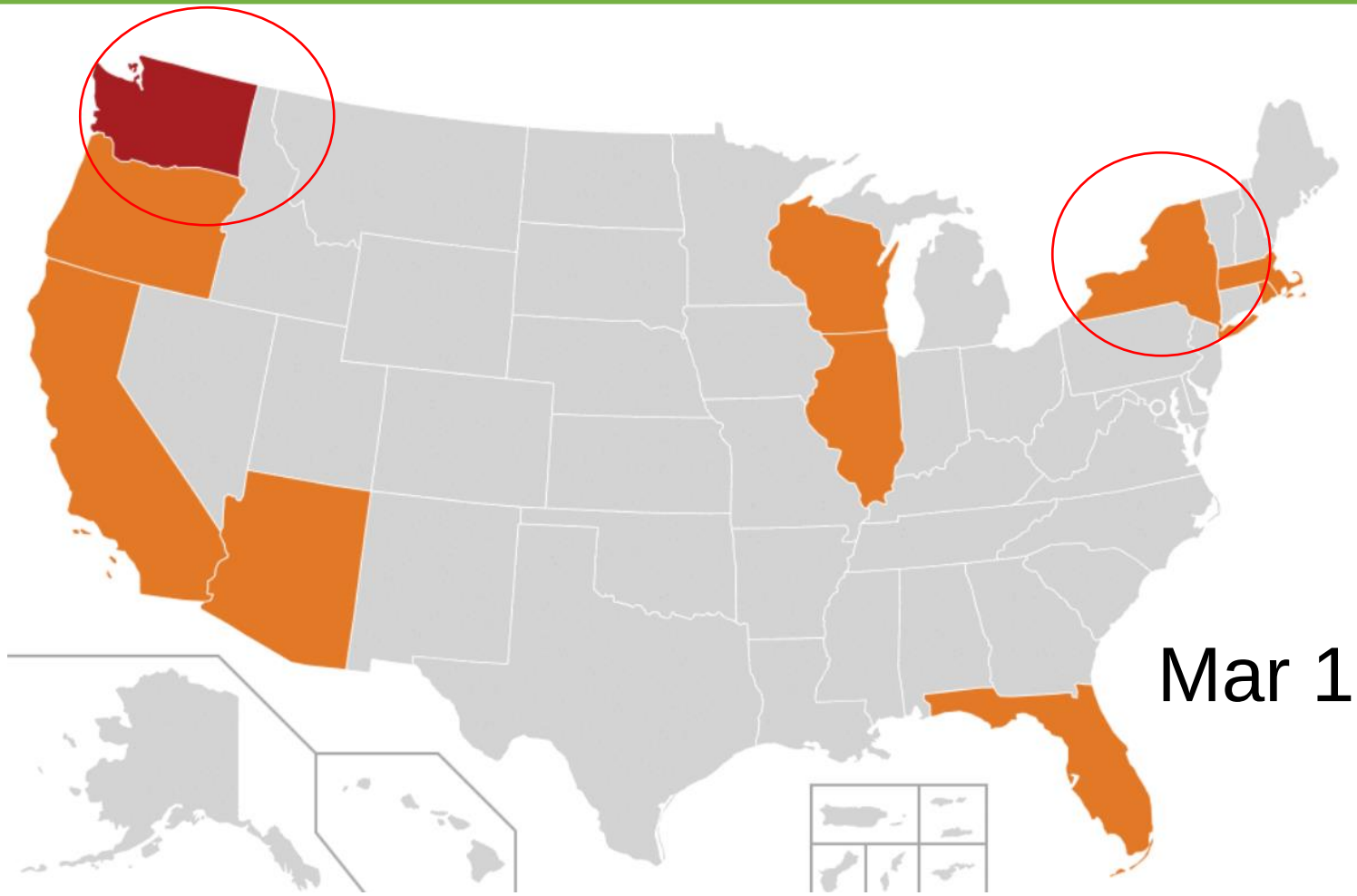
- ❑ Discuss the UAB Fungal Reference Lab COVID-19 experience
- ❑ Introduce the Thermo Fisher Ion Torrent Genexus Sequencing Platform and Ion AmpliSeq SARS-CoV-2 Insight Research Assay
- ❑ Discuss lab implementation/workflow
- ❑ Highlight the impact of our sequencing data on public health



JANUARY 21 - FIRST CASE OF SARS-COV-2 INFECTION REPORTED IN WASHINGTON STATE

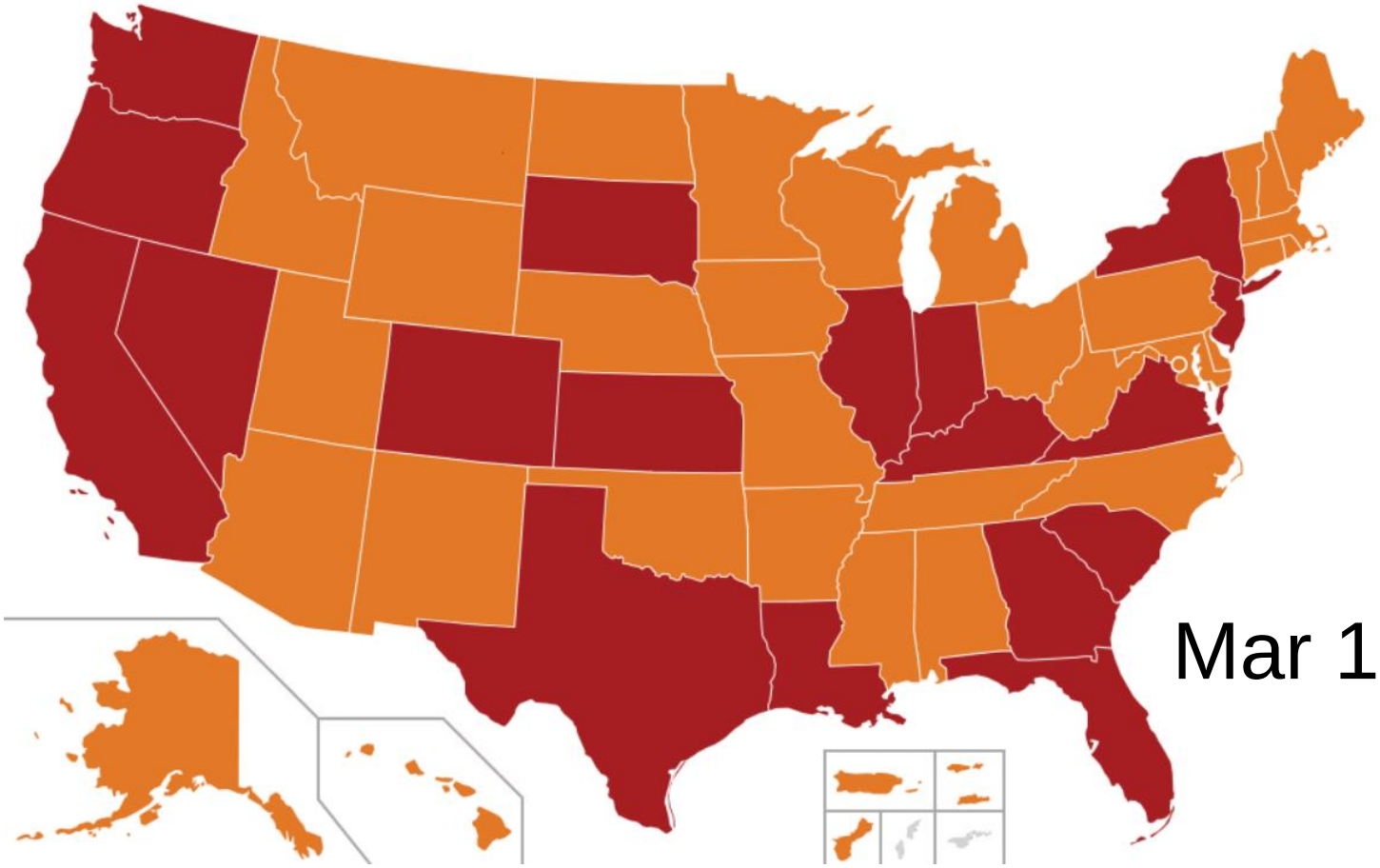


MARCH 1 - MAJORITY OF COMMERCIAL DIAGNOSTIC TESTS APPROPRIATED TO "HOT SPOTS" (WASHINGTON, NY)



Commercial tests NOT available. URGENT need to obtain reagents and develop a novel PCR assay

MARCH 17 - UAB FRL SARS-COV-2 TEST GOES LIVE



Mar 17

24 H TAT (MOST SAME DAY) ENABLED UAB HOSPITALS AND MANY REGIONAL HOSPITALS TO RETURN TO OPERATION

UAB

LEGACY of HOPE

Alabama's Organ & Tissue Donation Alliance

CORONER
DEPUTY
JEFFERSON COUNTY
MEDICAL EXAMINER

IN-HOUSE TESTING ENABLED THE START OF CLINICAL TRIALS AND BIOSPECIMEN REPOSITORY

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Center for Clinical and Translational Science

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Partner Network Research Commons Training Academy Clinical Translation Engagement of Communities Special Modules News & Events About

Plans to unite our students, faculty and staff and UAB's mission to fight COVID-19

- COVID-19 Enterprise Research Initiative
- Informatics
- BERD
- Panels
- Funding Opportunities
- Grant Help
- Research Implementation
- OnCore
- Commercialization
- Trainings
- Resources

Contact

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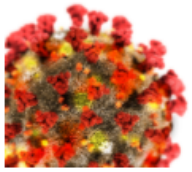
jcroker@uab.edu



COVID-19 Enterprise Research Initiative

Home Biospecimen Clinical Data Biomedical Data Teams & Working Groups Tools & Resources Literature

Contact

Working with the Division of Infectious Diseases as well as other UAB divisions and departments and coordinated by the CCTS, the IRB has endorsed an Enterprise Research Platform for COVID-19 to shepherd the safe and efficient consent process and collection of specimens and clinical data, which is ongoing. CCTS Informatics, with the Center for Outcomes & Effectiveness Research, Informatics Institute and multiple schools, is coordinating clinical data and longitudinal follow-up for rigorous secondary analysis that ensures efficient and respectful engagement of research participants. This new Enterprise Platform will serve to anchor and enable all COVID-19-related human subjects research across the campus.

Enterprise Advantages:

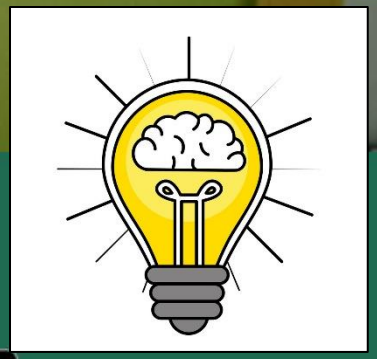
- Recruitment process that protects the safety and health of research teams and patients
- Coordinated strategy for specimen and data collection that respects the good will of the participant
- Consent for longitudinal follow-up, recall and invitation to participate in future studies

JUNE 22 - INITIATIVE TO TEST 250,000 COLLEGE STUDENTS PRIOR TO RETURN TO SCHOOL

GuideSafe™

Home Testing Tools Actions Impact Media About Support

The tools and resources to help fight COVID-19 and safely get back to life.



Protect Your Community. Protect Your Privacy. Protect Your Life. Stop the Spread of COVID in Alabama.

Get the App. Download now.

Download on the App Store

GET IT ON Google Play

JULY 28-AUG 30 – THE GUIDESAFE INITIATIVE TEAM RESULTED UP TO 11,337 TESTS PER DAY

- ❑ Helped implement a safe return to college campuses in the State of AL
- ❑ Resources and infrastructure subsequently used for sentinel testing

AUG 09 - SUNDAY NIGHT 2AM - “POOLING” AN ALL-NIGHTER

SARS-COV-2 EXPANSION FROM MARCH TO AUGUST

SARS-CoV-2 Expansion from March to August

~ 150 test per day → **Up to 11,000 test per day**

DECEMBER 2020 - REPORTS OF A NEW MORE INFECTIOUS UK VARIANT

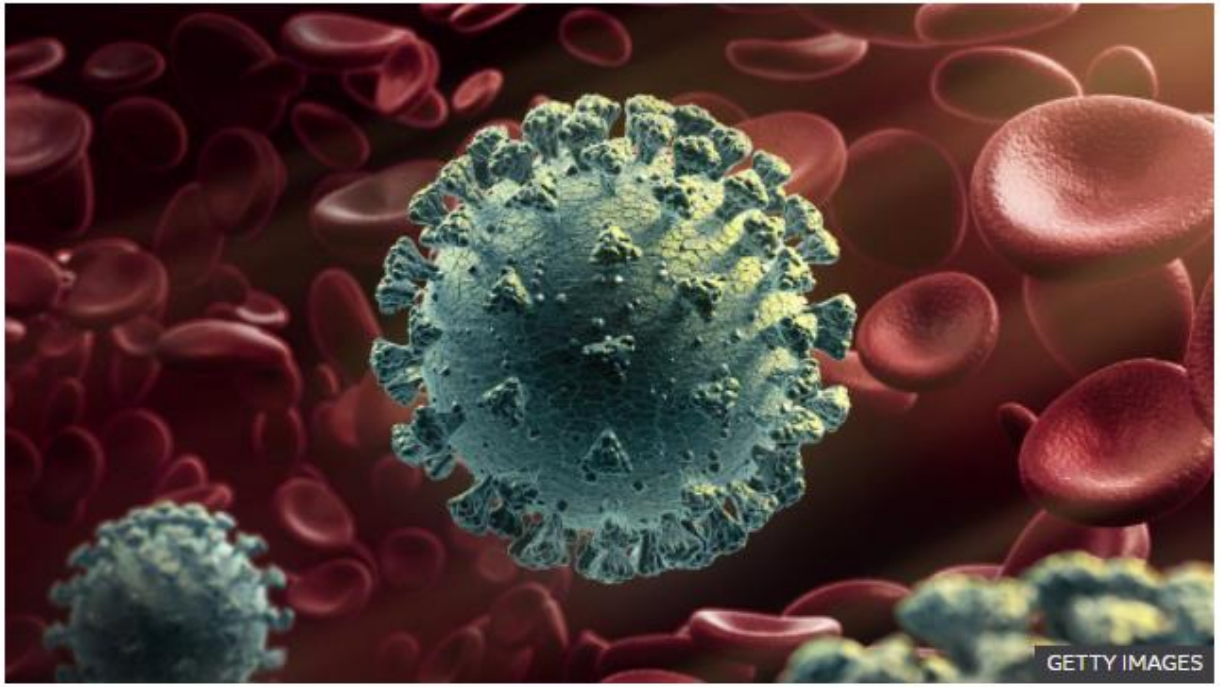
BBC NEWS

New coronavirus variant: What do we know?

By James Gallagher
Health and science correspondent

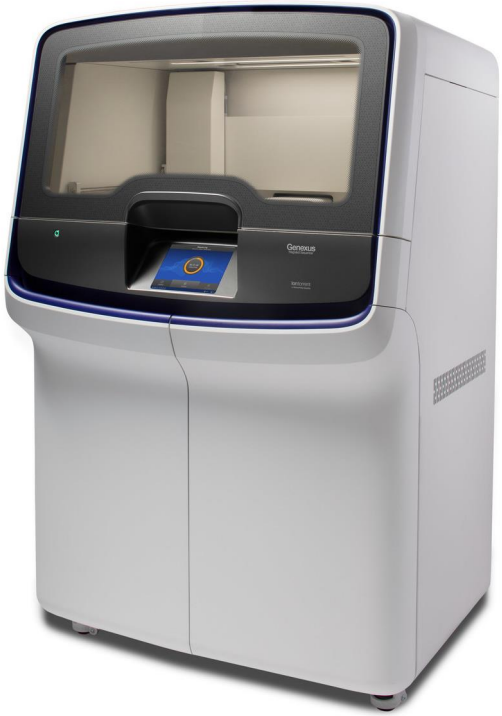
20 December 2020

Coronavirus pandemic

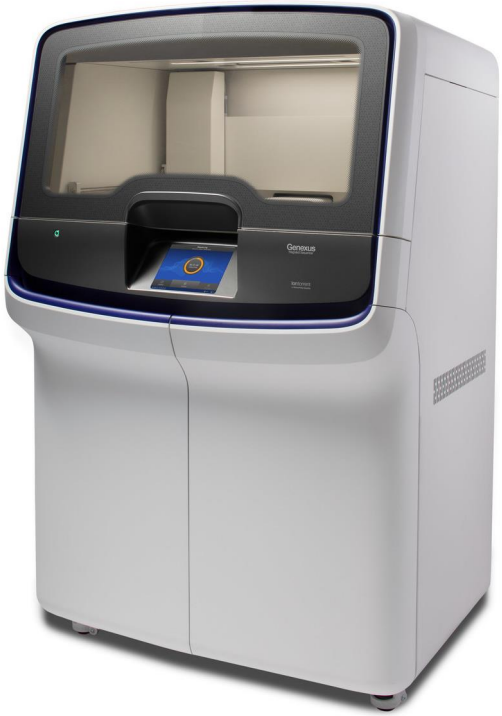


DECEMBER 2020- IMPLEMENTED THE ION AMPLISEQ SARS-COV-2 INSIGHT RESEARCH ASSAY ON 3 ION TORRENT GENEXUS SYSTEMS

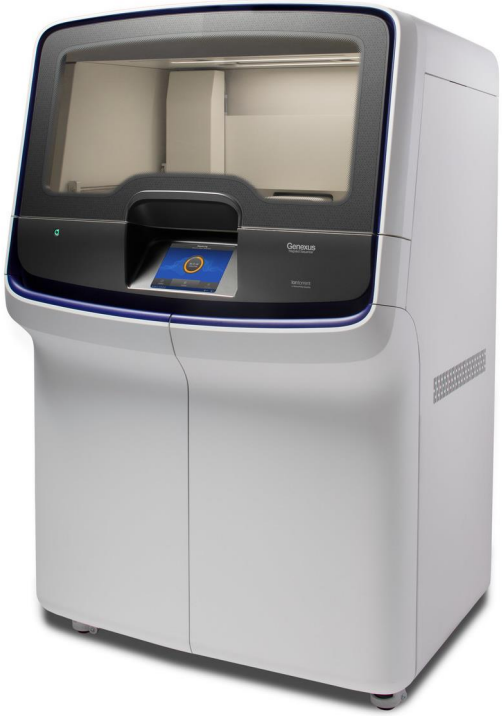
Deep Space Nine



Galileo



USS Enterprise



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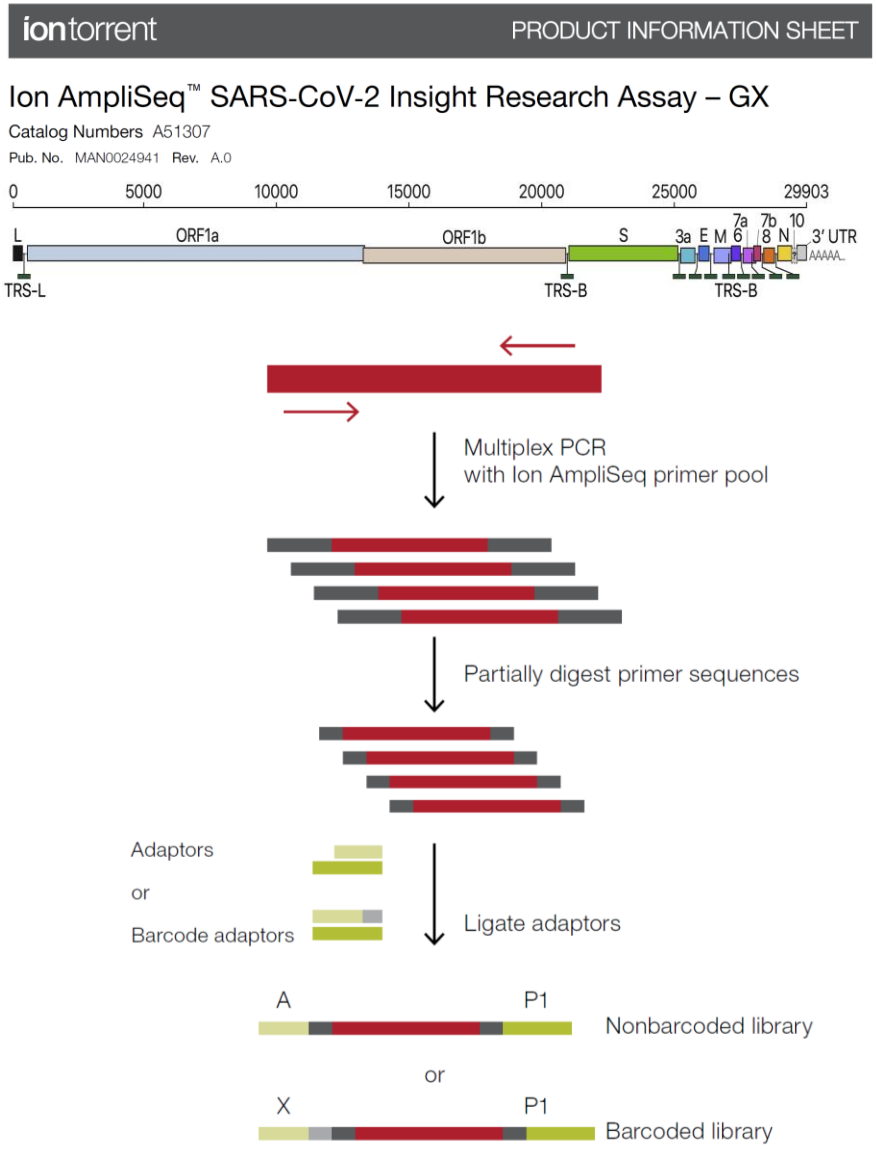
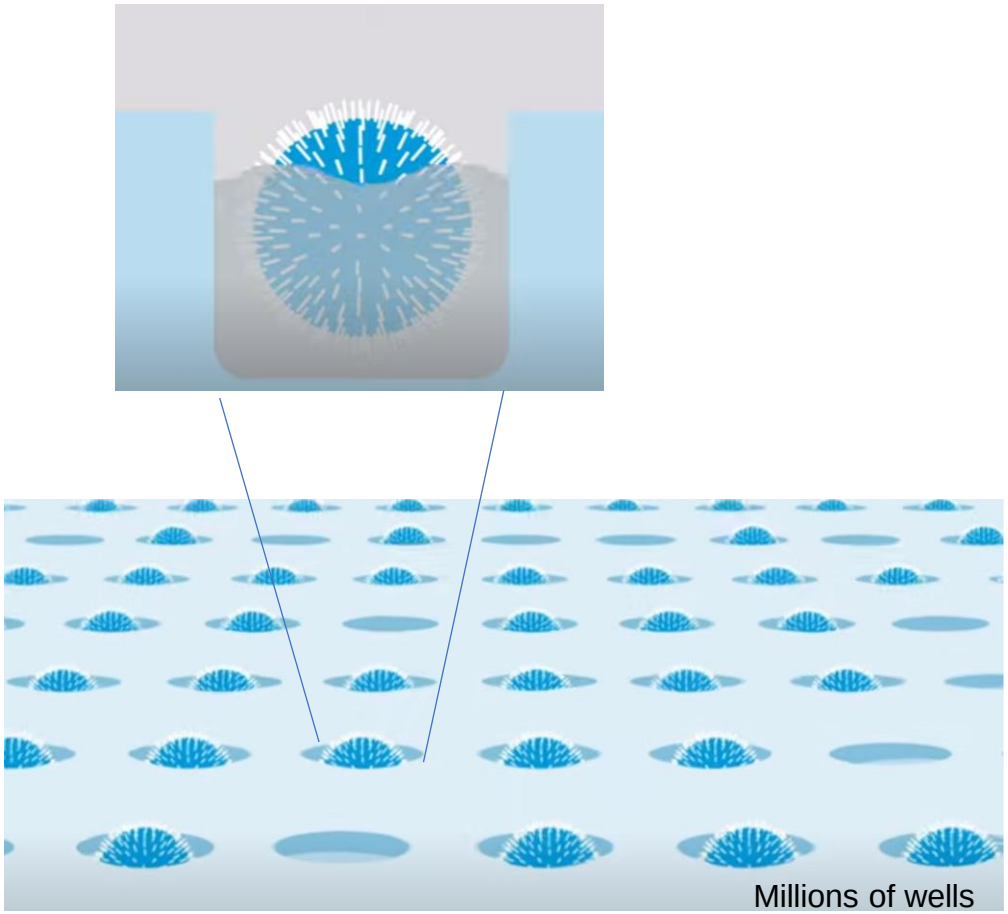
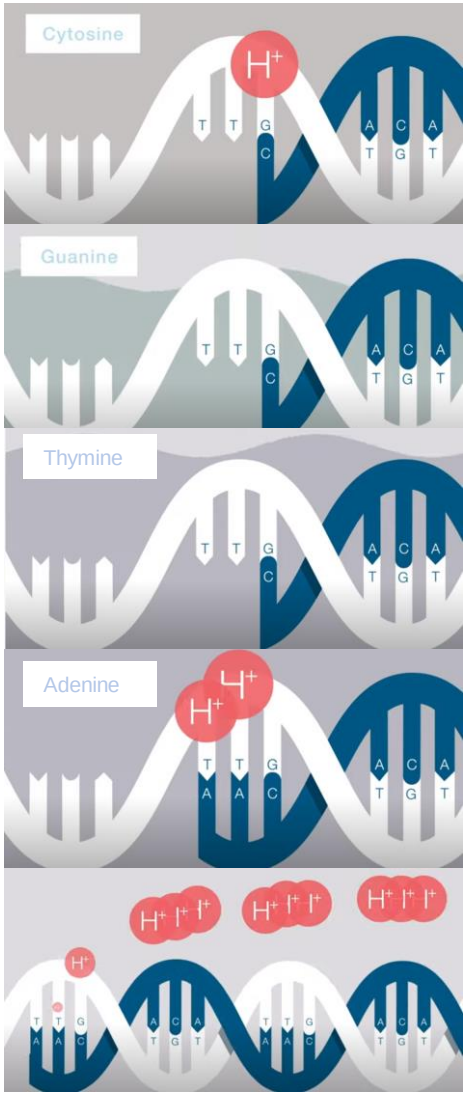
AMPLICON BASED TARGET ENRICHMENT
MULTIPLEX PCR→ ~ 180BP OVERLAPPING AMPLICONS WITH BARCODED ADAPTORS→ ENABLE > 99% GENOME COVERAGE



ION TORRENT - SYNTHESIS BY SEQUENCING

PCR EXPANDS SINGLE AMPLICON ON BEAD → BEAD IN WELL WITH PH METER→ NUCLEOTIDES ADDED SEQUENTIALLY → H+ RELEASED





WORKFLOW- EXTRACT, MEASURE, DILUTE RNA→ LOAD RNA/ BUFFERS/CONSUMABLES/SEQUENCING CHIP→ ~24H OBTAIN A LIST OF VARIANT CALLS & IRMA FASTA SEQUENCE

Start with viral RNA
Run setup: 5 min of hands-on time

Automate on Genexus Integrated Sequencer

cDNA synthesis
Initialization, sample dilution, and reverse transcription

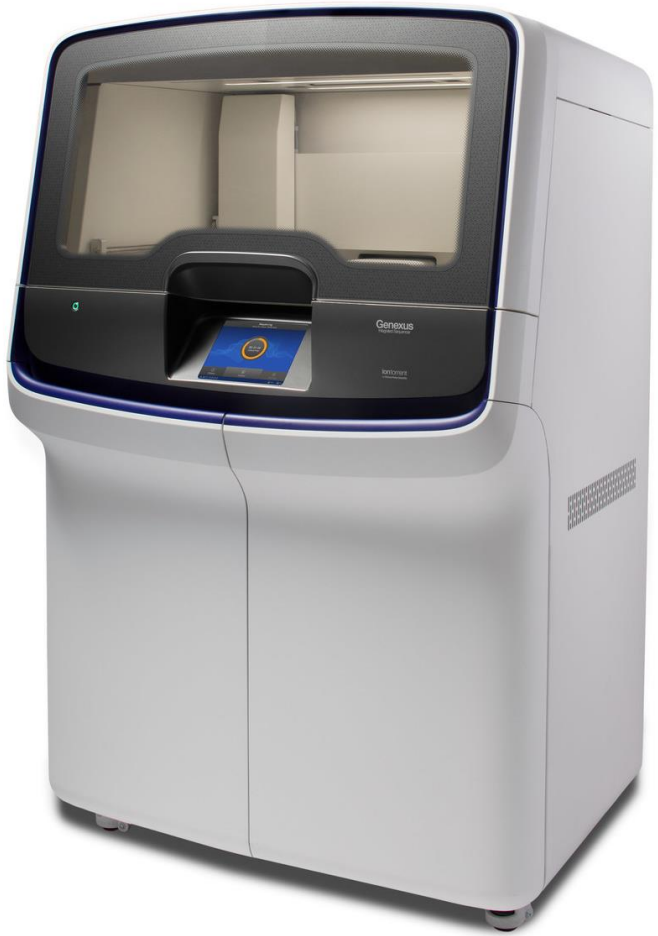
Library preparation
Ion AmpliSeq library preparation and library equalization

Template preparation
Amplification of library onto Ion Sphere Particles loaded onto GX5 Chip

Sequencing
Sequential flows of natural nucleotides measuring incorporation events

Post-run analysis
Base calling, variant calling using plug-ins: SnpEff, IRMA, Trinity

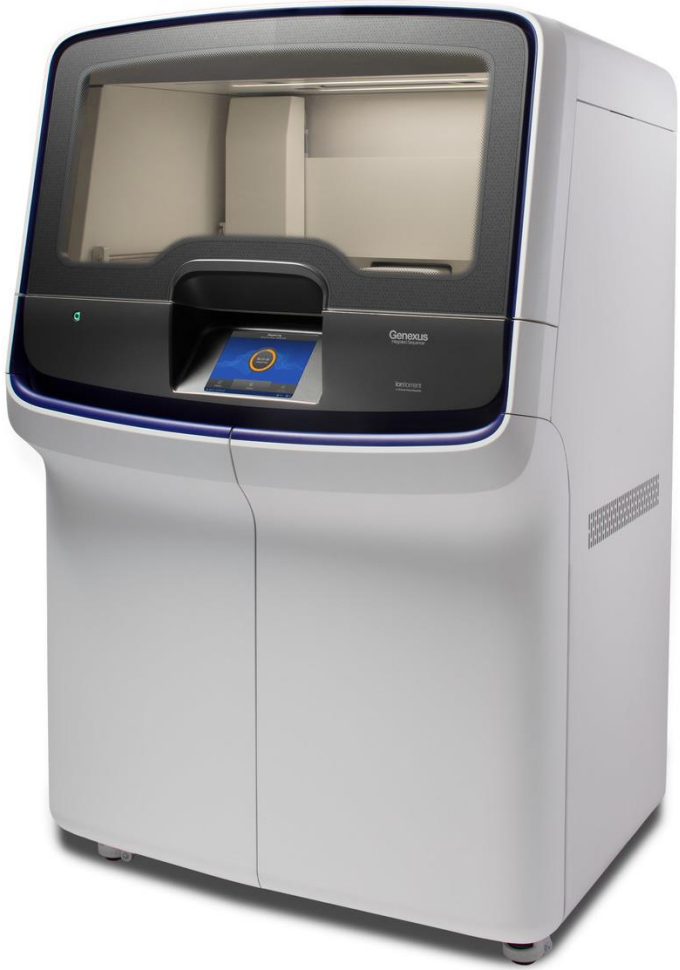
Get variant report in 24 hr



THE UNIVERSITY OF ALABAMA AT BIRMINGHAM

WORKFLOW- EXTRACT, MEASURE, DILUTE RNA→ LOAD RNA/ BUFFERS/CONSUMABLES/SEQUENCING CHIP→ ~24H OBTAIN A LIST OF VARIANT CALLS & IRMA FASTA SEQUENCE

1. Log samples
2. Confirm that viral quantity in the sample meets criteria
    1. Cycle threshold </= 30
3. Extract, Quantify, Dilute RNA
    1. ProMega Maxwell RSC48 system with viral RNA extraction kit
    2. ProMega Quantus fluorometer- measure RNA concentration-
    3. Genexus software assists with sample dilution factors
4. Load RNA in hood into PCR plate
    1. 1st 2 columns of plate (n=16)
    2. Seal with aluminum foil
5. Load Genexus system with the following:
    1. Sealed RNA plate
    2. 2 empty PCR plates
    3. Pipette Tips
    4. Consumable reagent strips 1-4
    5. Primer pool tubes
    6. Bar coding nucleotide tubes
    7. Sequencing chip
6. Close lid of system
7. Robotic arm preps reactions
    1. 1 preparation cycle→ 16 samples
8. Onboard thermocycler performs PCR
9. Amplicons on beads are loaded into the chip
    1. 1 chip→ 32 sequences total



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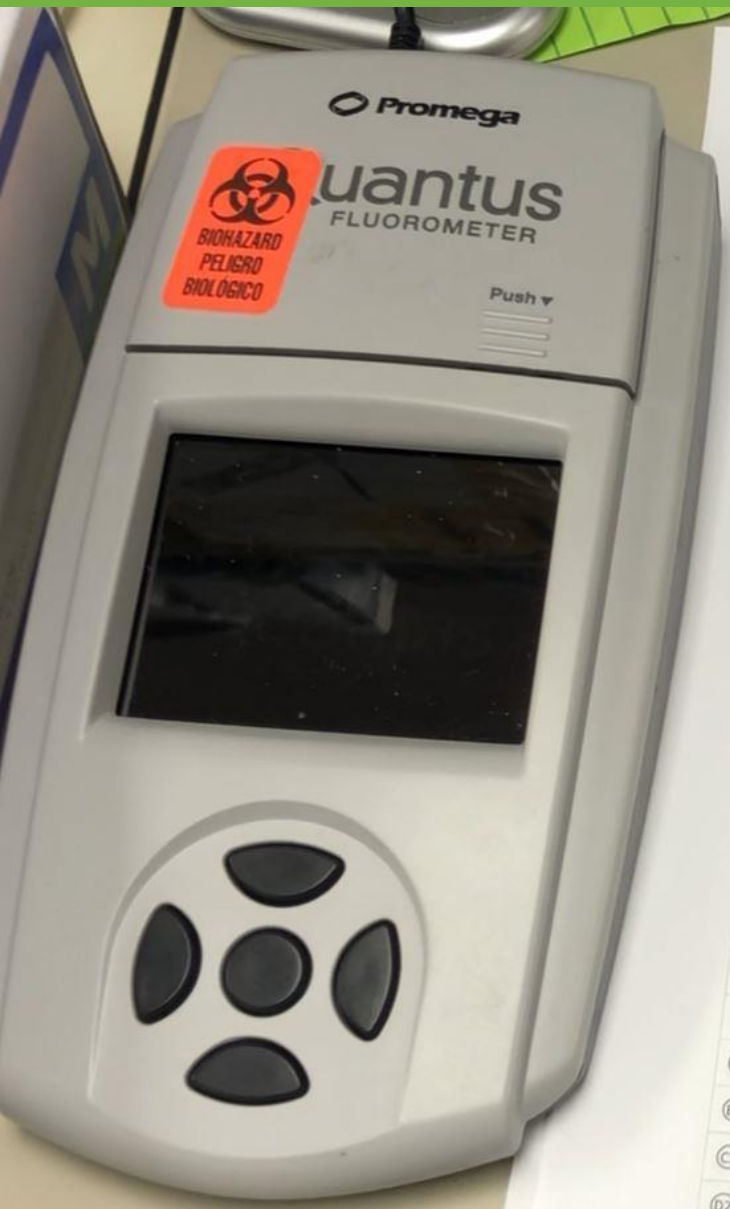
LOG SAMPLES; CONFIRM THAT CT VALUES MEET THE FRL VIRAL CONCENTRATION THRESHOLD SET AT </= 30

	A	B	C	D	E	F	G	H	I	J	K	L
1	FRL ID	First Name	Last Name	SeqID	Sample Date	Hospital	City, State	MRN	DOB	Sex	PCR platform/Ct/RLU	Tissue
1587	GX1586			GX1586								
1588	GX1587			GX1587								
1589	GX1588			GX1588								
1590	GX1589			GX1589								
1591	GX1590			GX1590								
1592	GX1591			GX1591								
1593	GX1592			GX1592								
1594	GX1593			GX1593								
1595	GX1594			GX1594								
1596	GX1595			GX1595								
1597	GX1596			GX1596								
1598	GX1597			GX1597								

EXTRACT VIRAL RNA FROM THE NASAL SWAB

DETERMINE THE RNA CONCENTRATION BY FLUOROMETRY AND DILUTE THE SAMPLE PRIOR TO LOADING ONTO GENEXUS



Run Name: 23SEP21
Ion Reporter Software: N/A
Report Template: Default Report
Print This Page

Consumables Required For This Run

(1) Genexus™ Coupler
(8) Genexus™ Strip 1
(3) 96 Well Plates
(2) Genexus™ Bottle 2
(1) Genexus™ Bottle 3
(1) GXS™ Chip
(8) Genexus™ Strip 2-AS
(2) Genexus™ Strip 3-GXS™
(1) Genexus™ Bottle 1
(1) Ion Torrent™ Genexus™ Barcodes 1-96 AS - 32 barcodes will be used from this plate
(4) Ion Torrent™ Genexus™ Pipette tips
(2) Genexus™ Strip 4
(1) Genexus™ Cartridge

Consumables Installed Ready To Use

None

Primer Tube Positions

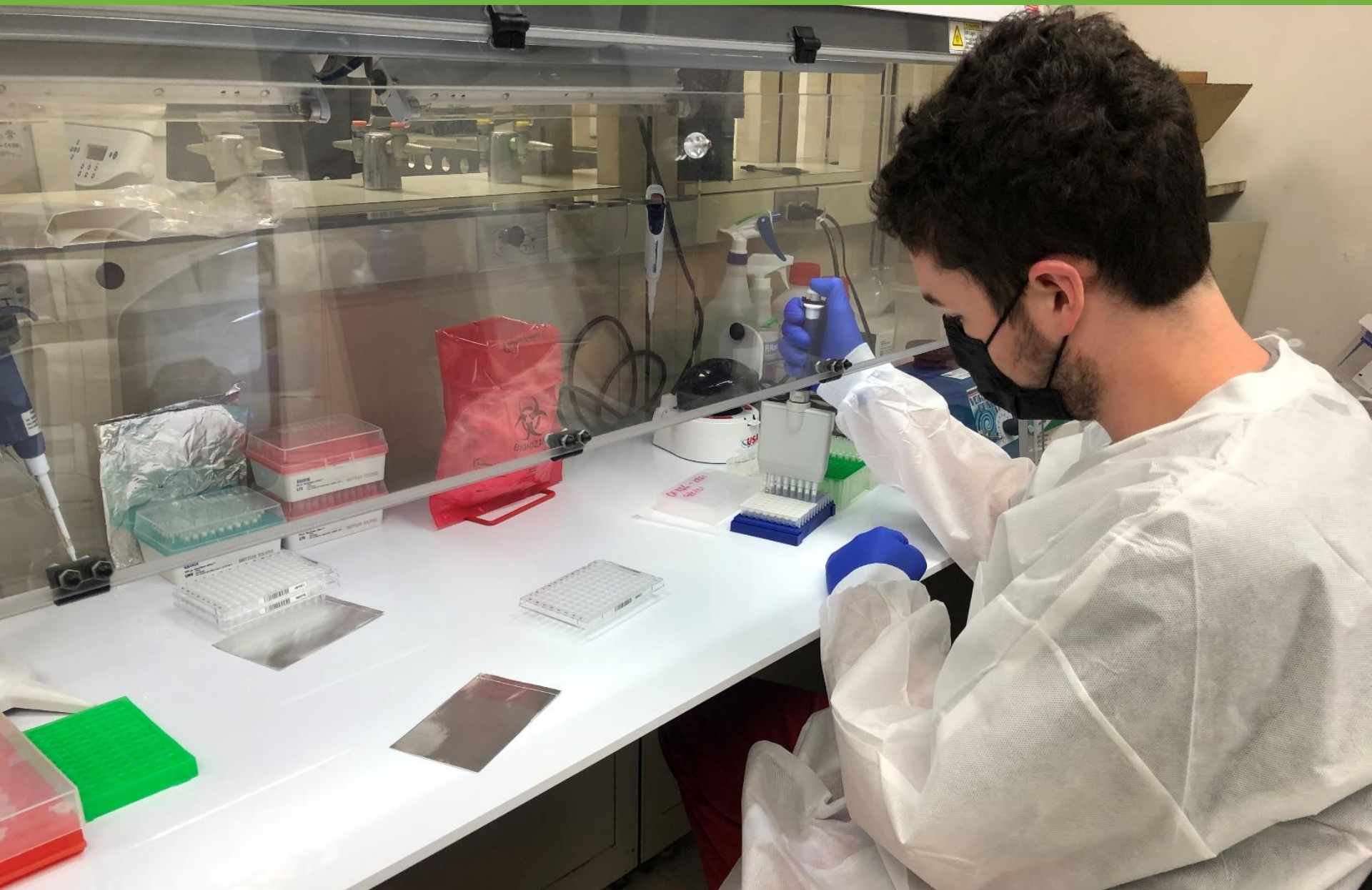
1 2 3 4 5 6 7 8

1. SARS-CoV-2 w2.0 [Place Genexus Primer Pool Tube 1 - RNA]
2. SARS-CoV-2 w2.0 [Place Genexus Primer Pool Tube 2 - RNA]
3. SARS-CoV-2 w2.0 [Place Genexus Primer Pool Tube 1 - RNA]
4. SARS-CoV-2 w2.0 [Place Genexus Primer Pool Tube 2 - RNA]
5. SARS-CoV-2 w2.0 [Place Genexus Primer Pool Tube 1 - RNA]
6. SARS-CoV-2 w2.0 [Place Genexus Primer Pool Tube 2 - RNA]
7. SARS-CoV-2 w2.0 [Place Genexus Primer Pool Tube 1 - RNA]
8. SARS-CoV-2 w2.0 [Place Genexus Primer Pool Tube 2 - RNA]

Sample Locations

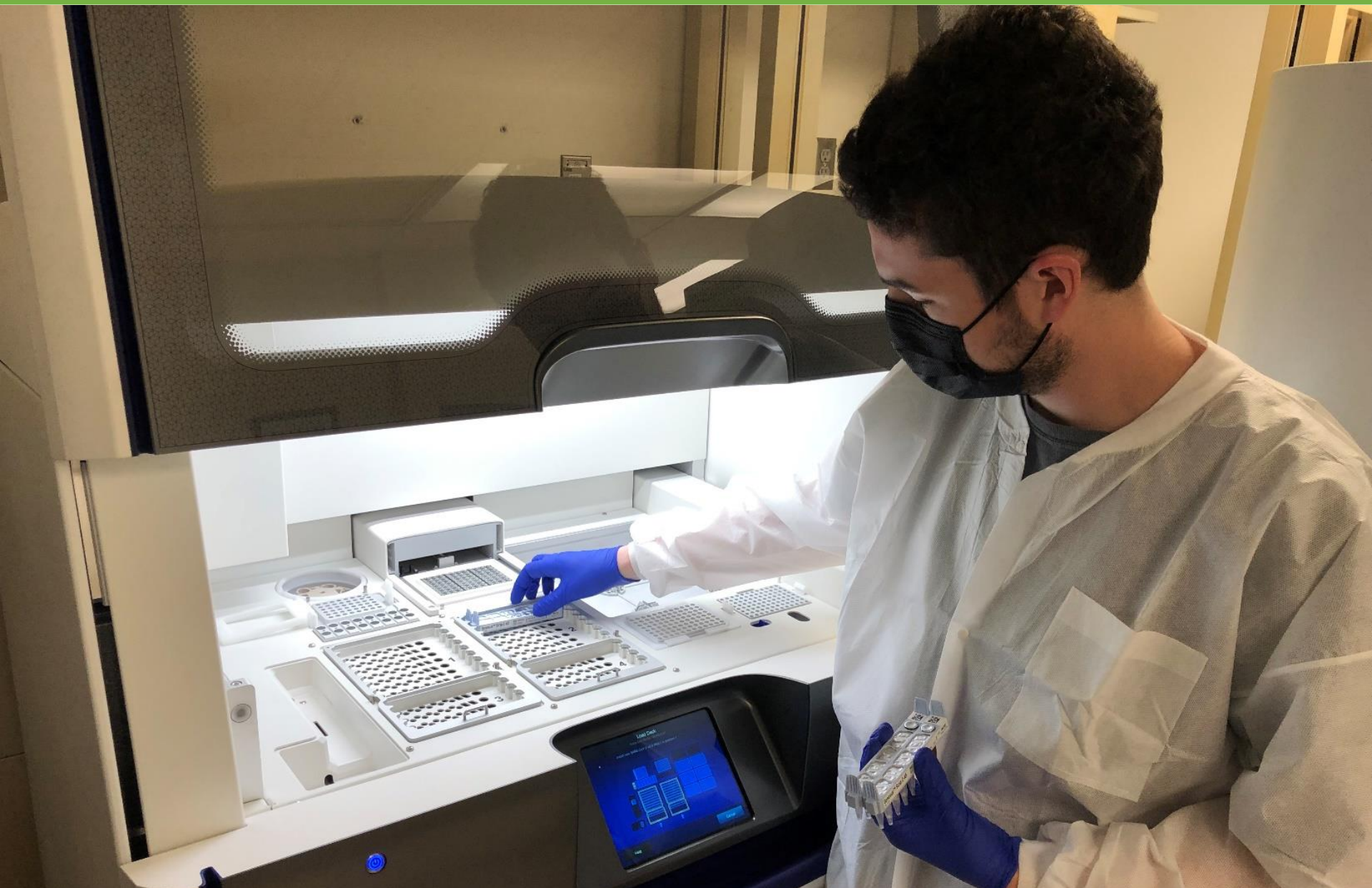
Well Pos.	Sample Name	Nucleic Acid T...	Vol. (µl)	Conc.(n...	Dilution Fac...	Assay Short Name
A1	GX1214	RNA	25.0	16	66.67	SARS-CoV-2 w2.0
B1	GX1215	RNA	25.0	18	75	SARS-CoV-2 w2.0
C1	GX1216	RNA	25.0	17	70.83	SARS-CoV-2 w2.0
D1	GX1217	RNA	25.0	16	66.67	SARS-CoV-2 w2.0
E1	GX1218	RNA	25.0	20	83.33	SARS-CoV-2 w2.0
F1	GX1219	RNA	25.0	17	70.83	SARS-CoV-2 w2.0
G1	GX1220	RNA	25.0	19	79.17	SARS-CoV-2 w2.0
H1	GX1221	RNA	25.0	23	95.83	SARS-CoV-2 w2.0
A2	GX1222	RNA	25.0	20	83.33	SARS-CoV-2 w2.0
B2	GX1223	RNA	25.0	55.3	230.42	SARS-CoV-2 w2.0
C2	GX1224	RNA	25.0	19	79.17	SARS-CoV-2 w2.0
D2	GX1225	RNA	25.0	132		

TRANSFER RNA INTO 1ST TWO COLUMNS OF A PCR PLATE AND SEAL WITH ALUMINUM FOIL



LOAD NEW BUFFERS/WASH FLUIDS INTO THE GENEXUS SYSTEM

LOAD TEST STRIPS 1-4 INTO THE GENEXUS SYSTEM

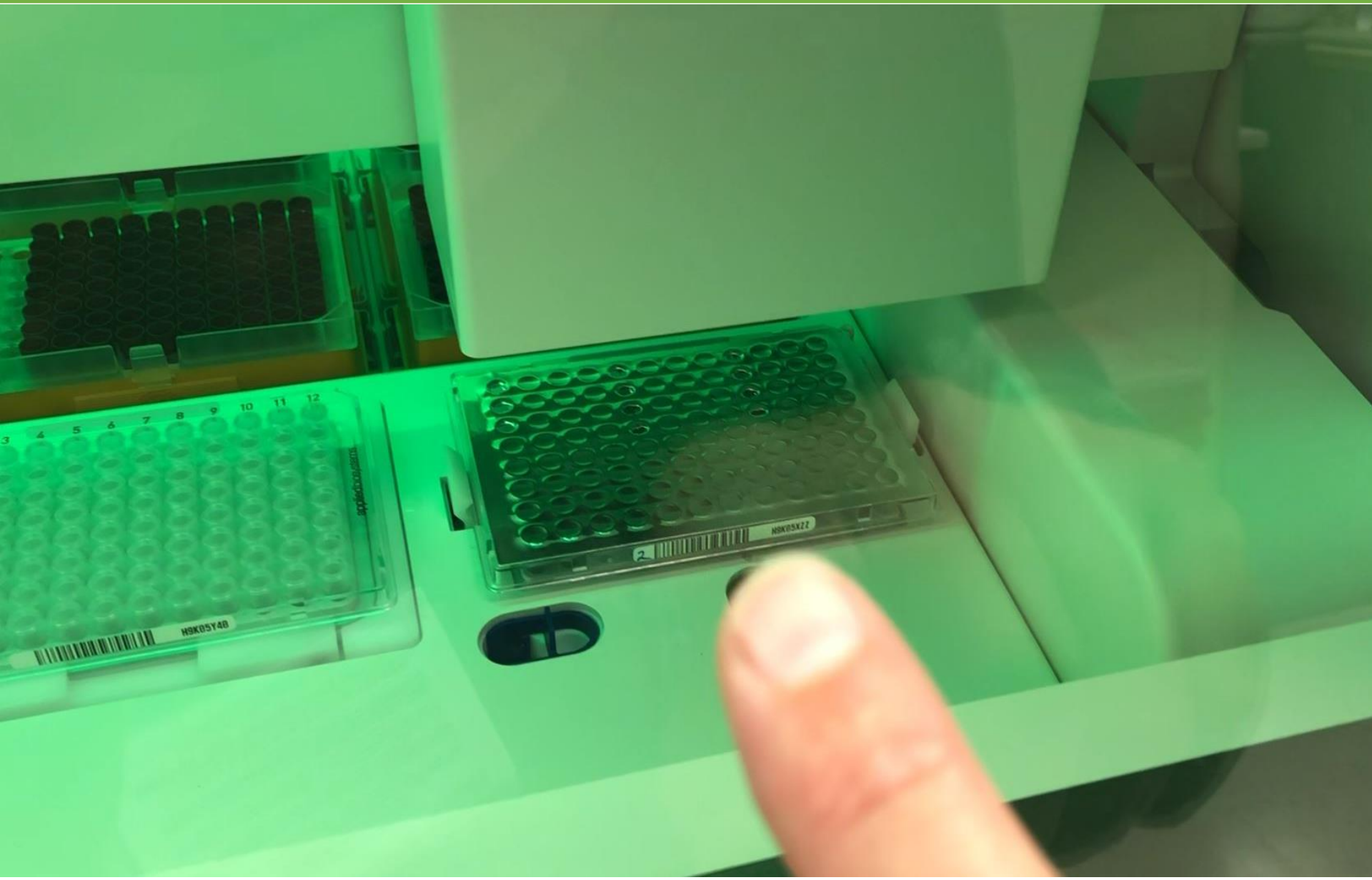


LOAD TEST STRIPS 1-4 INTO THE GENEXUS SYSTEM

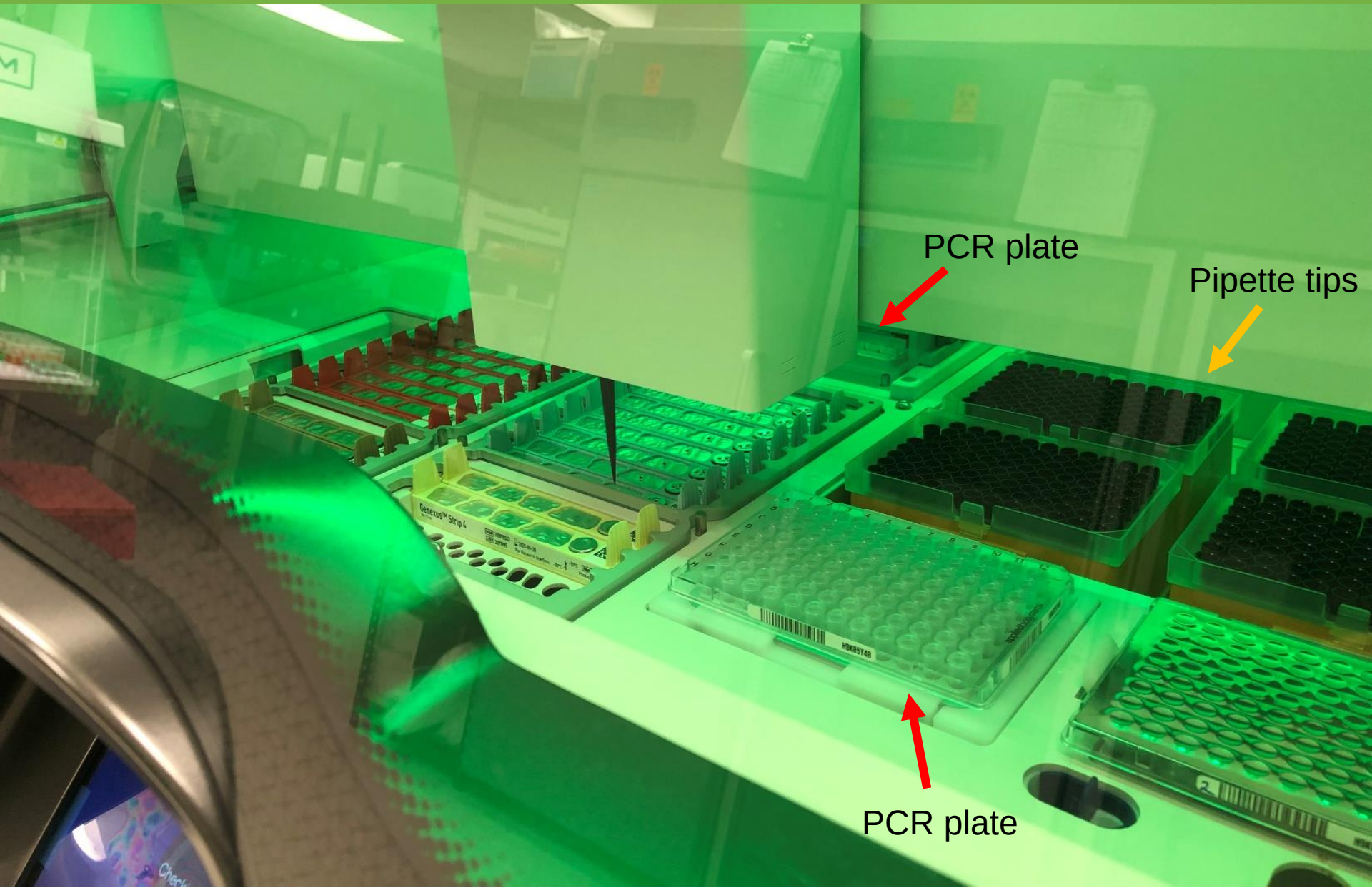
LOAD THE SEQUENCING CHIP INTO THE GENEXUS SYSTEM

LOAD TUBES WITH PRIMER POOLS INTO THE GENEXUS SYSTEM

LOAD THE RNA PLATE INTO THE GENEXUS SYSTEM



LOAD PIPETTE TIPS AND 2 EMPTY PCR PLATES INTO THE GENEXUS SYSTEM



REPORT- VARIANT CALL LIST

SAMPLE REPORT

Sequencing Lab Report : Ion Ampliseq SARS-CoV-2-LowTiter Research Assay 2.0

	A	B	C	D	E	F	G	H	I	J	K	L	M	N	O	P	Q
1	CHROM	POS	REF	ALT	VARTYPE	GEN[*].GT	ANN[*].GENE	ANN[*].FEATUREID	EFF[*].RANK	ANN[*].HGVS_C	ANN[*].HGVS_P	ANN[*].EFFECT	EFF[*].IMPACT	AF	QUAL	DP	SSSB
2	2019-nCoV	241	C	T	SNP	Homozygous	orf1ab	QHD43415.1	-1	c.-25C>T	.	upstream_gene_variant	MODIFIER	0.999498	14578.1	6883	2.16E-04
3	2019-nCoV	913	C	T	SNP	Homozygous	orf1ab	QHD43415.1	1	c.648C>T	p.Ser216Ser	synonymous_variant	LOW	0.996	14552.7	6673	4.60E-05
4	2019-nCoV	2110	C	T	SNP	Homozygous	orf1ab	QHD43415.1	1	c.1845C>T	p.Asn615Asn	synonymous_variant	LOW	0.997999	14673.1	12103	0.00103053
5	2019-nCoV	3037	C	T	SNP	Homozygous	orf1ab	QHD43415.1	1	c.2772C>T	p.Phe924Phe	synonymous_variant	LOW	0.995468	14658.9	23338	-6.47E-04
6	2019-nCoV	3267	C	T	SNP	Homozygous	orf1ab	QHD43415.1	1	c.3002C>T	p.Thr1001Ile	missense_variant	MODERATE	0.995477	14453.8	2659	5.33E-04
7	2019-nCoV	5388	C	A	SNP	Homozygous	orf1ab	QHD43415.1	1	c.5123C>A	p.Ala1708Asp	missense_variant	MODERATE	1	14831.6	6585	3.42E-04
8	2019-nCoV	5986	C	T	SNP	Homozygous	orf1ab	QHD43415.1	1	c.5721C>T	p.Phe1907Phe	synonymous_variant	LOW	0.998999	14609.9	4416	5.39E-04
9	2019-nCoV	6954	T	C	SNP	Homozygous	orf1ab	QHD43415.1	1	c.6689T>C	p.Ile2230Thr	missense_variant	MODERATE	0.997997	14711.9	12614	4.98E-04
10	2019-nCoV	7984	T	C	SNP	Homozygous	orf1ab	QHD43415.1	1	c.7719T>C	p.Asp2573Asp	synonymous_variant	LOW	0.997997	14722.2	18490	5.21E-06
11	2019-nCoV	10747	C	T	SNP	Homozygous	orf1ab	QHD43415.1	1	c.10482C>T	p.Asn3494Asn	synonymous_variant	LOW	0.987481	14139.6	18195	-0.00441907
12	2019-nCoV	11287	GTCTGGTTTT	G	DEL	Homozygous	orf1ab	QHD43415.1	1	c.11023_11031delTCTGGTTTT	p.Ser3675_Phe3677del	conservative_inframe_deletion	MODERATE	1	14367	16905	0
13	2019-nCoV	14120	C	T	SNP	Homozygous	orf1ab	QHD43415.1	2	c.13856C>T	p.Pro4619Leu	missense_variant	MODERATE	0.991988	14380.3	20530	-0.00395635
14	2019-nCoV	14408	C	T	SNP	Homozygous	orf1ab	QHD43415.1	2	c.14144C>T	p.Pro4715Leu	missense_variant	MODERATE	0.986987	14039.3	5013	0.00132514
15	2019-nCoV	14676	C	T	SNP	Homozygous	orf1ab	QHD43415.1	2	c.14412C>T	p.Pro4804Pro	synonymous_variant	LOW	0.99198	14307.5	18552	-0.00100984
16	2019-nCoV	15279	C	T	SNP	Homozygous	orf1ab	QHD43415.1	2	c.15015C>T	p.His5005His	synonymous_variant	LOW	0.9965	14604.4	4035	2.25E-04
17	2019-nCoV	16176	T	C	SNP	Homozygous	orf1ab	QHD43415.1	2	c.15912T>C	p.Thr5304Thr	synonymous_variant	LOW	0.993994	14468.7	11035	-3.81E-04
18	2019-nCoV	17004	C	T	SNP	Homozygous	orf1ab	QHD43415.1	2	c.16740C>T	p.Leu5580Leu	synonymous_variant	LOW	0.998999	14806.2	7560	8.31E-04
19	2019-nCoV	17518	C	T	SNP	Homozygous	orf1ab	QHD43415.1	2	c.17254C>T	p.Leu5752Phe	missense_variant	MODERATE	0.998997	14708.6	17282	-7.26E-05
20	2019-nCoV	19390	C	T	SNP	Homozygous	orf1ab	QHD43415.1	2	c.19126C>T	p.Pro6376Ser	missense_variant	MODERATE	0.991976	14298	5310	-3.94E-04
21	2019-nCoV	21764	ATACATG	A	DEL	Homozygous	S	QHD43416.1	1	c.204_209delACATGT	p.His69_Val70del	disruptive_inframe_deletion	MODERATE	1	14500.6	9668	3.99E-08
22	2019-nCoV	21990	TTTA	T	DEL	Homozygous	S	QHD43416.1	1	c.432_434delTTA	p.Tyr145del	disruptive_inframe_deletion	MODERATE	0.998978	14391.6	6275	0
23	2019-nCoV	23063	A	T	SNP	Homozygous	S	QHD43416.1	1	c.1501A>T	p.Asn501Tyr	missense_variant	MODERATE	0.995484	14458.1	4347	2.86E-04
24	2019-nCoV	23271	C	A	SNP	Homozygous	S	QHD43416.1	1	c.1709C>A	p.Ala570Asp	missense_variant	MODERATE	0.998997	14757.6	12017	8.35E-05
25	2019-nCoV	23403	A	G	SNP	Homozygous	S	QHD43416.1	1	c.1841A>G	p.Asp614Gly	missense_variant	MODERATE	0.998492	14571.9	33490	-0.00173152
26	2019-nCoV	23604	C	A	SNP	Homozygous	S	QHD43416.1	1	c.2042C>A	p.Pro681His	missense_variant	MODERATE	0.994478	14428	29192	0.00128508
27	2019-nCoV	23709	C	T	SNP	Homozygous	S	QHD43416.1	1	c.2147C>T	p.Thr716Ile	missense_variant	MODERATE	0.998997	14708.5	24333	4.44E-04
28	2019-nCoV	24506	T	G	SNP	Homozygous	S	QHD43416.1	1	c.2944T>G	p.Ser982Ala	missense_variant	MODERATE	0.995992	14496.5	24385	-0.00196254
29	2019-nCoV	24914	G	C	SNP	Homozygous	S	QHD43416.1	1	c.3352G>C	p.Asp1118His	missense_variant	MODERATE	0.995491	14471.3	11205	0.00275755
30	2019-nCoV	27972	C	T	SNP	Homozygous	ORF8	QHD43422.1	1	c.79C>T	p.Gln27*	stop_gained	HIGH	0.998999	14788	21009	3.66E-04
31	2019-nCoV	28048	G	T	SNP	Homozygous	ORF8	QHD43422.1	1	c.155G>T	p.Arg52Ile	missense_variant	MODERATE	0.996986	14600.7	21287	-3.34E-04
32	2019-nCoV	28095	A	T	SNP	Homozygous	ORF8	QHD43422.1	1	c.202A>T	p.Lys68*	stop_gained	HIGH	0.996491	14262.1	9136	-0.00818679
33	2019-nCoV	28111	A	G	SNP	Homozygous	ORF8	QHD43422.1	1	c.218A>G	p.Tyr73Cys	missense_variant	MODERATE	0.998996	14710.2	9209	9.05E-04
34	2019-nCoV	28280	GAT	CTA	MNP	Homozygous	N	QHD43423.2	1	c.7_9delGATinsCTA	p.Asp3Leu	missense_variant	MODERATE	1	14341.3	13057	0
35	2019-nCoV	28881	GGG	AAC	MNP	Homozygous	N	QHD43423.2	1	c.608_610delGGGinsAAC	p.ArgGly203LysArg	missense_variant	MODERATE	0.99949	14551	3344	-3.31E-04
36	2019-nCoV	28977	C	T	SNP	Homozygous	N	QHD43423.2	1	c.704C>T	p.Ser235Phe	missense_variant	MODERATE	0.999499	14779	3391	9.13E-04

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REPORT- IRMA FASTA CONSENSUS SEQUENCE

SAMPLE REPORT

Sequencing Lab Report : Ion Ampliseq SARS-CoV-2-LowTiter Research Assay 2.0

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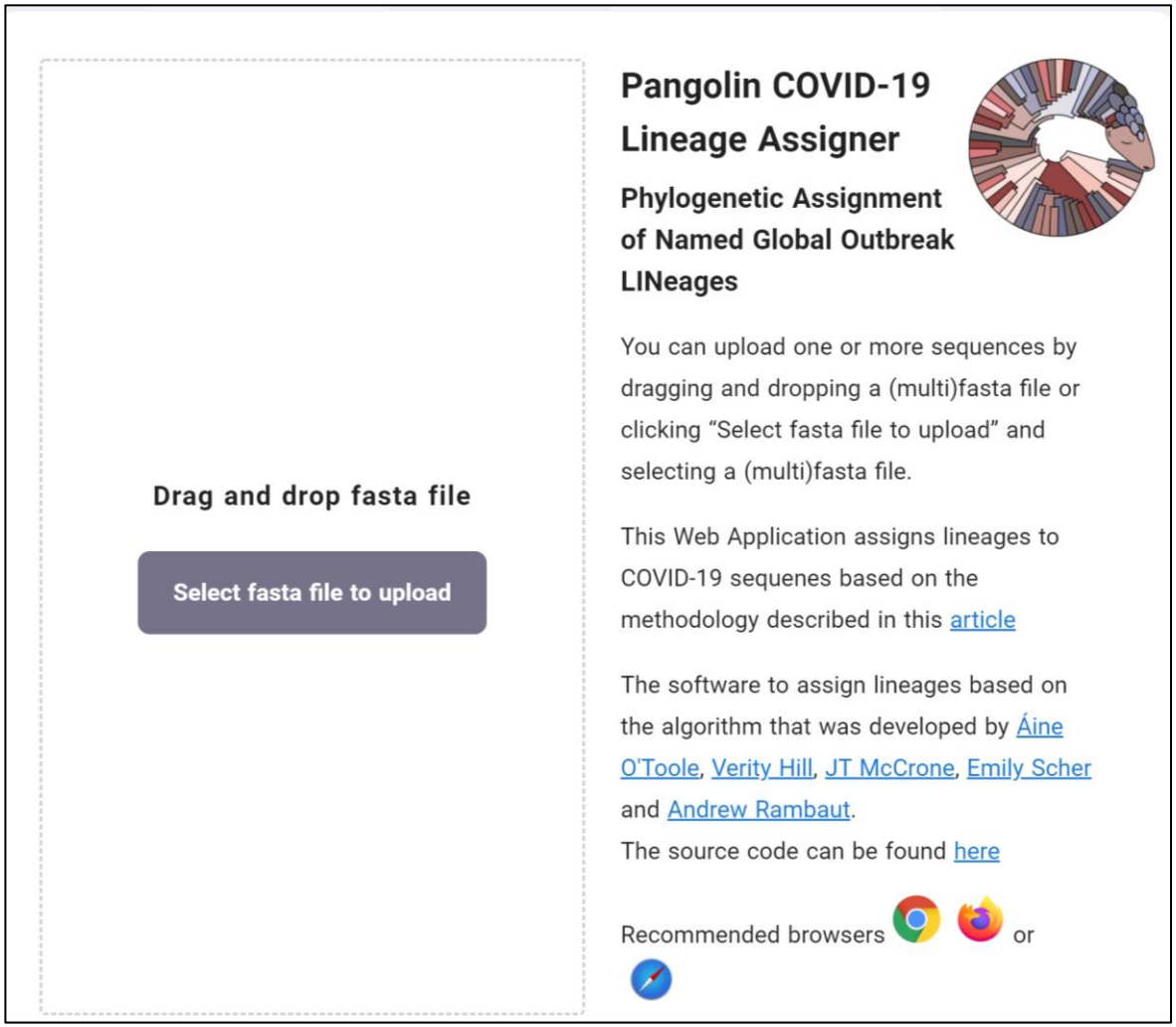
UPLOAD IRMA FASTA CONSENSUS FILE INTO PANGOLIN COVID-19 LINEAGE ASSIGNER PROGRAM TO DETERMINE LINEAGE



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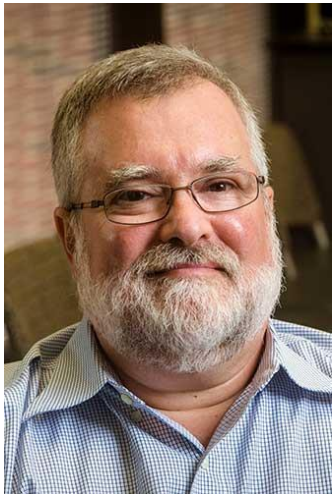
WE INCORPORATED INDEPENDENT CONFIRMATION OF CONSENSUS SEQUENCES AND LINEAGE CALLS



Elliot Lefkowitz

Curtis Hendrickson

THE UNIVERSITY OF ALABAMA AT BIRMINGHAM

REPORT RESULTS TO THE AL DEPARTMENT OF PUBLIC HEALTH

ALABAMA PUBLIC HEALTH
Promoting, protecting, and improving Alabama's health

	A	B	C	D	E	F	G	H	I	J	K	L	M	N
1	**FRL ID**	**First Name**	**Last Name**	**SeqID**	**Sample Date**	**Hospital**	**MRN**	**DOB**	**Sex**	**Date Sequenced**	**IRMA contig**	**Lineage: Pangolin of IRMA assembly**	**Signout**	**NCBI Accession #**
2					1/25/2021					1/26/2021	29,843	17/17 B.1.1.7 SNPs	B.1.1.7	MW651022
3					1/25/2021					1/26/2021	29,856	17/17 B.1.1.7 SNPs	B.1.1.7	MW651023
4					1/26/2021					1/28/2021	29,754	17/17 B.1.1.7 SNPs	B.1.1.7	MW651024
5					1/26/2021					1/28/2021	29,844	17/17 B.1.1.7 SNPs	B.1.1.7	MW651025
6					1/27/2021					1/28/2021	29,869	17/17 B.1.1.7 SNPs	B.1.1.7	MW651026
7					1/24/2021					1/26/2021	29,849	16/17 B.1.1.7 SNPs	B.1.1.7	MW651027
8					1/26/2021					1/28/2021	29830	B.1.2	B.1.2	MW651028
9					1/26/2021					1/28/2021	29848	B.1.521	B.1.521	MW653862

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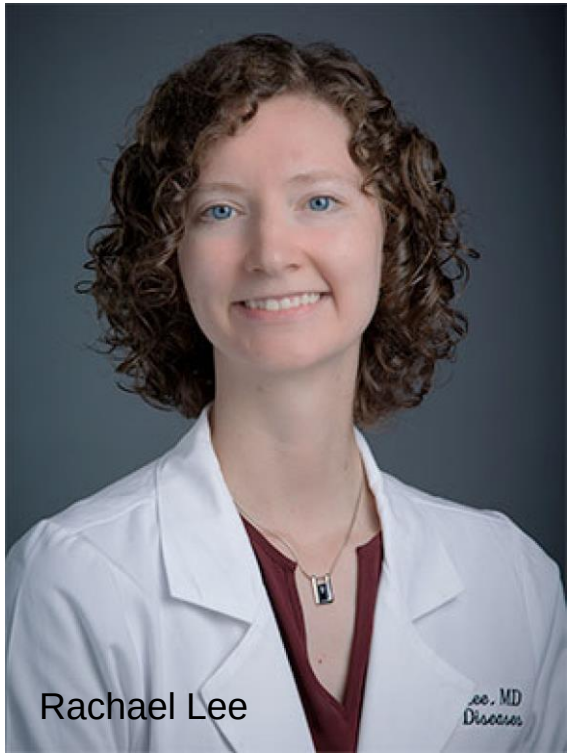
REPORT RESULTS TO UAB INFECTION PREVENTION



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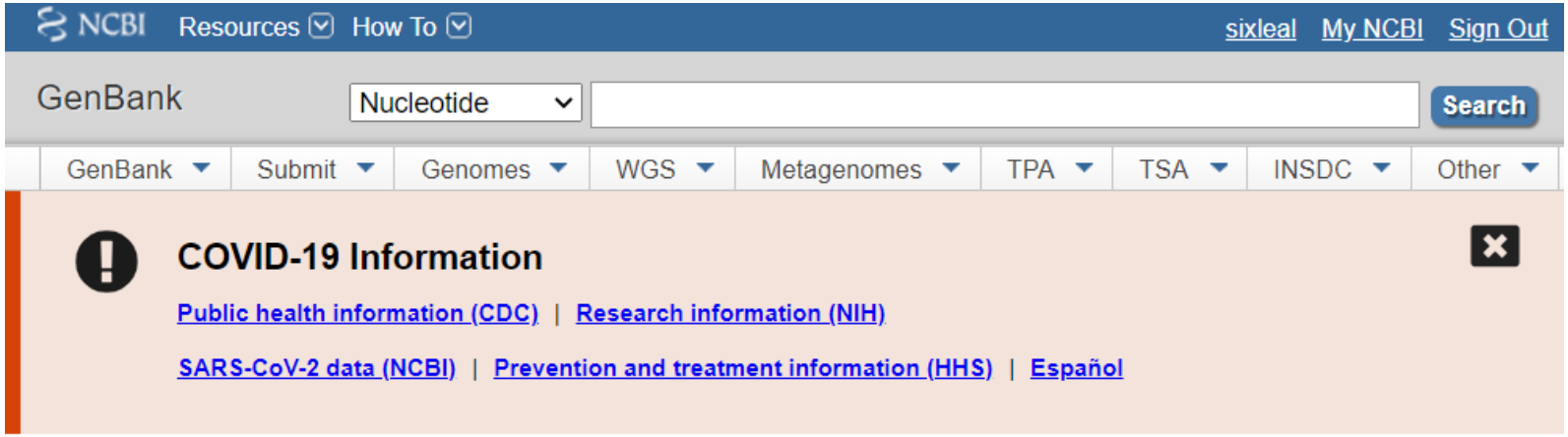
UPLOAD FASTA FILES TO NCBI GENBANK



THE UNIVERSITY OF ALABAMA AT BIRMINGHAM

UPLOAD FASTA FILES TO GISAID

THE UNIVERSITY OF ALABAMA AT BIRMINGHAM

AS OF 9/1, OUR TEAM HAS SEQUENCED A TOTAL OF 890 SAMPLES

Samples Sequenced prior to 9/1: 890

Sequences available within 3-4 days of samples collection

Max Num of Sequences per instrument per month: 320, this assumes that you run 16 samples/run and you complete one run each day Mon-Fri.

Monthly Breakdown of Samples:

	# Sequenced	# Failed	Total Sequenced	% Successful
Jan	9	1	8	88.89
Feb	54	7	47	87.04
Mar	62	12	50	80.65
Apr	122	17	105	86.07
May	85	6	79	92.94
Jun	48	3	45	93.75
Jul	241	6	235	97.51
Aug	336	15	321	95.54

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OUR TEAM WAS THE 1ST IN THE STATE OF AL TO DETECT ALL MAJOR VARIANTS OF PUBLIC HEALTH SIGNIFICANCE

Monthly Breakdown by Variant:										
	Jan	Feb	Mar	Apr	May	Jun	Jul	Aug	Sep	
B.1.1.7 (alpha) "UK"	6	42	39	78	55	15	10	2		247
B.1.2	1	1	5	5	2	1	0	0		15
B.1.521	1	0	0	0	0	0	0	0		1
B.1	0	1	0	0	1	1	0	0		3
B.1.234	0	2	0	0	0	0	0	0		2
B.1.1.222	0	1	0	0	0	0	0	0		1
B.1.1.519	0	0	3	2	0	0	0	0		5
B.1.1.1	0	0	1	4	0	1	0	0		6
B.1.427 (Epsilon) "NY"	0	0	0	1	0	0	0	0		1
B.1.351 (Beta) "South Africa"	0	0	0	1	0	0	0	0		1
B.1.526.2	0	0	0	1	0	0	0	0		1
B.1.1.306	0	0	0	2	0	0	0	0		2
B.1.525 (Eta) "CA"	0	0	0	1	0	0	0	0		1
B.1.540	0	0	0	0	1	0	0	0		1
B.1.596	0	0	0	2	0	0	0	0		2
B.1.526 (Iota) "CA"	0	0	2	1	1	2	0	0		6
R.1	0	0	0	1	0	0	0	0		1
B.1.1.28.1	0	0	0	0	0	0	0	0		0
C.37	0	0	0	2	16	5	0	0		23
P.1 (Gamma) "Brazil"	0	0	0	1	3	3	3	3		13
P.1.7	0	0	0	0	0	0	0	1		1
B.1.621 (Mu) "Peru"	0	0	0	0	0	1	0	0		1
B.1.626	0	0	0	0	0	0	0	0		0
B.1.627	0	0	0	0	0	0	1	0		1
B.1.623	0	0	0	0	0	2	5	0		7
B.1.617.2 (Delta) "Indian"	0	0	0	1	0	14	189	269		473
AY.1 (Delta) "Indian"	0	0	0	0	0	0	0	0		0
AY.2 (Delta) "Indian"	0	0	0	0	0	0	1	1		2
AY.3 (Delta)"Indian"	0	0	0	0	0	0	26	31		57
AY.3.1 (Delta) "Indian"	0	0	0	0	0	0	0	11		11
AY.4 (Delta) "Indian"	0	0	0	0	0	0	0	3		3
AY.5 (Delta) "Indian"	0	0	0	0	0	0	0	0		0
AY.14 (Delta) "Indian"	0	0	0	0	0	0	0	0		0
AY.25 (Delta) "Indian"	0	0	0	0	0	0	0	0		0
Totals/Month	8	47	50	103	79	45	235	321	0	888

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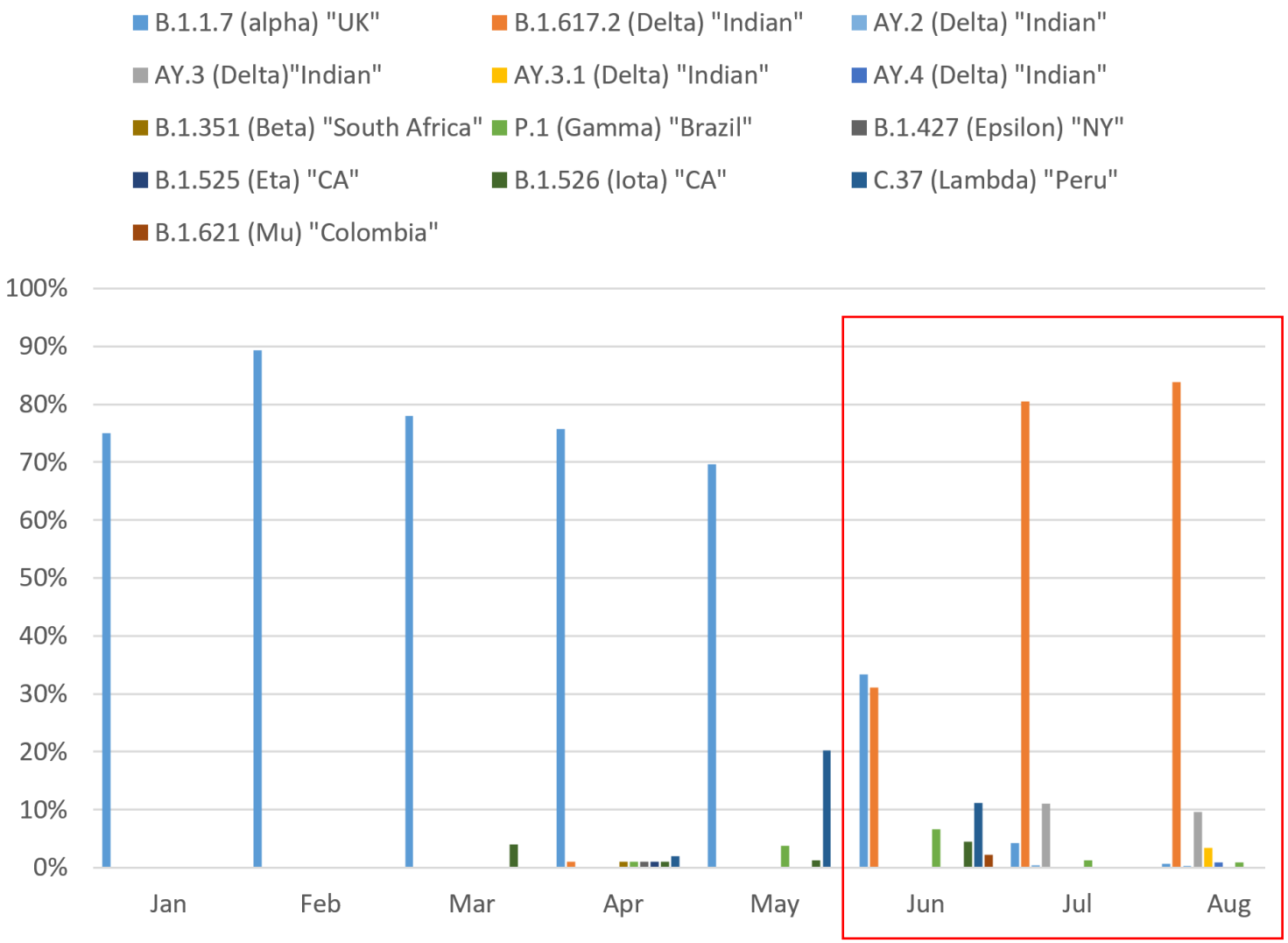
OUR TEAM'S DATA SHOWS ALPHA'S PREDOMINANCE FROM JAN-MAY AND DELTA'S RISE STARTING IN JUNE 2021



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LOW VACCINATION RATES WITH CONTINUED SELECTIVE PRESSURE BEG THE QUESTION: WHAT IS COMING NEXT?

Vaccinations by location

From Our World in Data · Last updated: 2 days ago

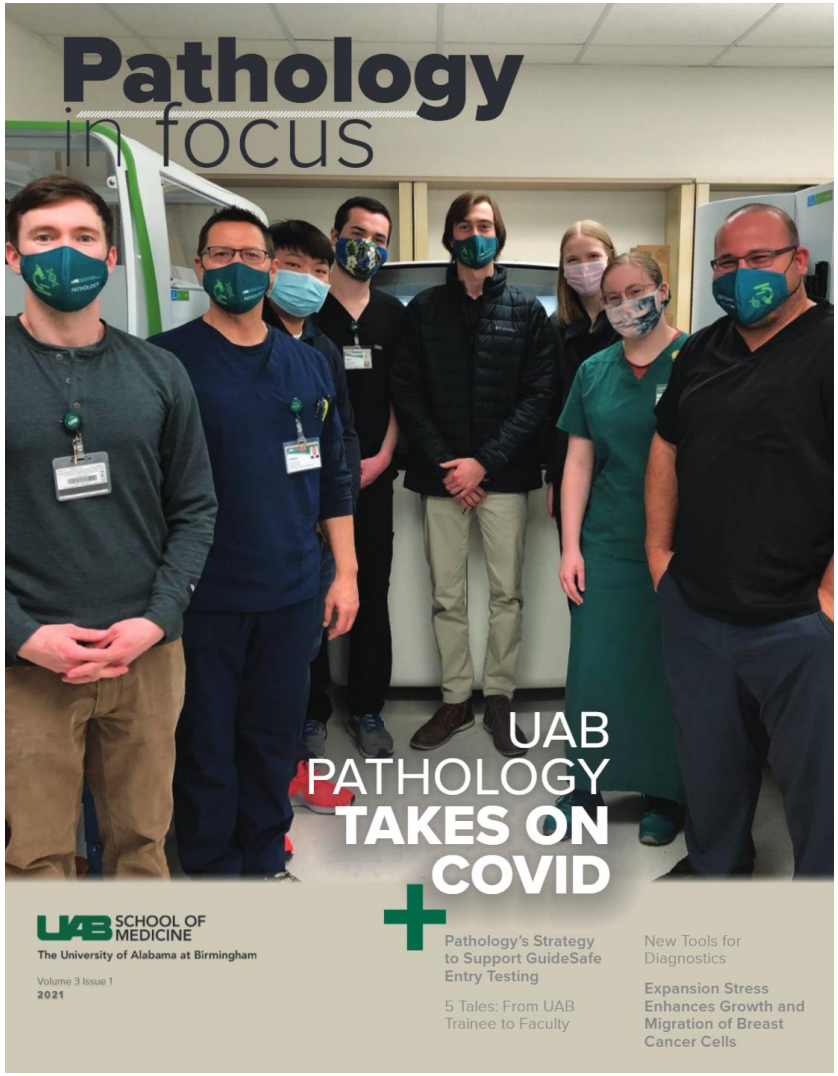
United States | All regions

Doses given	Fully vaccinated	% of population fully vaccinated
388M	183M	55.6%
+328K	+199K	+0.1%

Location	Doses given	Fully vaccinated	% of population fully vaccinated ↑
West Virginia	1.54M	721K	40.2%
Wyoming	512K	237K	41.0%
Idaho	1.56M	734K	41.1%
Alabama	4.44M	2.05M	41.7%
Mississippi	2.73M	1.27M	42.6%
U.S. Virgin Islands	101K	45,959	43.1%
North Dakota	732K	332K	43.5%
Georgia	10.5M	4.74M	44.6%
Tennessee	6.76M	3.05M	44.7%
Louisiana	4.45M	2.08M	44.8%
Arkansas	3.03M	1.36M	45.0%
South Carolina	5.16M	2.39M	46.4%
Oklahoma	4.08M	1.86M	46.9%
Missouri	6.2M	2.9M	47.3%
Indiana	6.66M	3.23M	48.0%
Montana	1.09M	514K	48.1%
North Carolina	11.1M	5.14M	49.0%

What viral variant is coming next?

OUR TEAM WILL CONTINUE TO WORK ON THE FRONTLINE TO DETECT VIRAL VARIANTS OF PUBLIC HEALTH SIGNIFICANCE IN AN EFFORT TO MITIGATE THE CURRENT PANDEMIC



Thank you for your time and attention

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