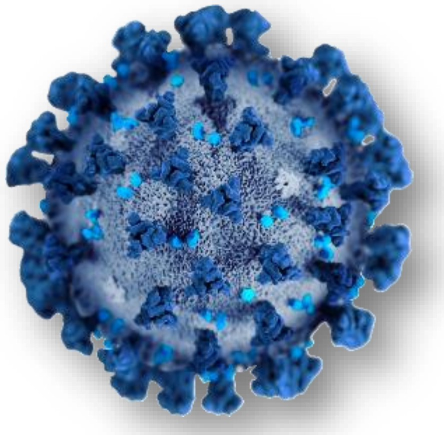


Pivoting from Cancer to COVID-19 in a global pandemic

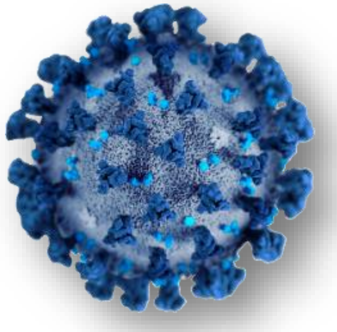
Paul Hofman, MD, PhD

UNIVERSITÉ CÔTE D'AZUR

Laboratory of Clinical and Experimental Pathology
Louis Pasteur Hospital, Nice, France
www.oncoage.org

OncoAge

Disclaimer



The COVID-19 crisis in Nice, France, and the impact in a pathology lab

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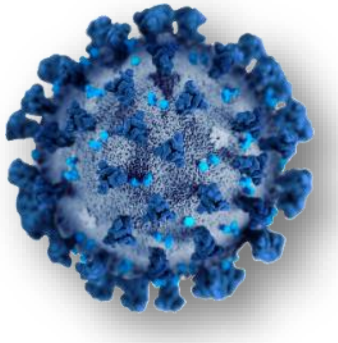

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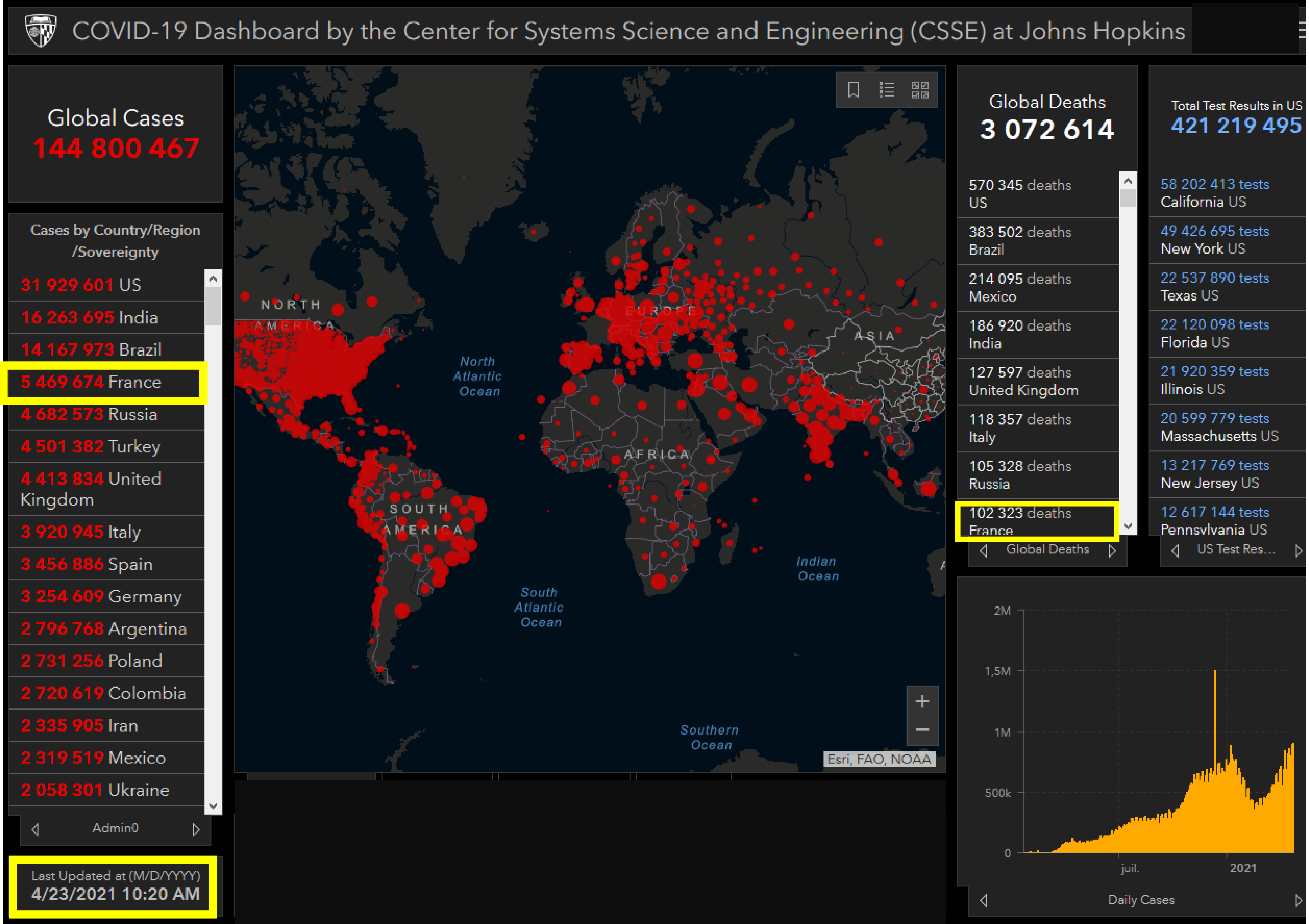


COVID-19 Dashboard by the Center for Systems Science and Engineering (CSSE) at Johns Hopkins

Global Cases
144 800 467

Cases by Country/Region/Sovereignty

31 929 601	US
16 263 695	India
14 167 973	Brazil
5 469 674	France
4 682 573	Russia
4 501 382	Turkey
4 413 834	United Kingdom
3 920 945	Italy
3 456 886	Spain
3 254 609	Germany
2 796 768	Argentina
2 731 256	Poland
2 720 619	Colombia
2 335 905	Iran
2 319 519	Mexico
2 058 301	Ukraine

Admin0

Last Updated at (M/D/YYYY)
4/23/2021 10:20 AM

Global Deaths
3 072 614

570 345 deaths	US
383 502 deaths	Brazil
214 095 deaths	Mexico
186 920 deaths	India
127 597 deaths	United Kingdom
118 357 deaths	Italy
105 328 deaths	Russia
102 323 deaths	France

Global Deaths

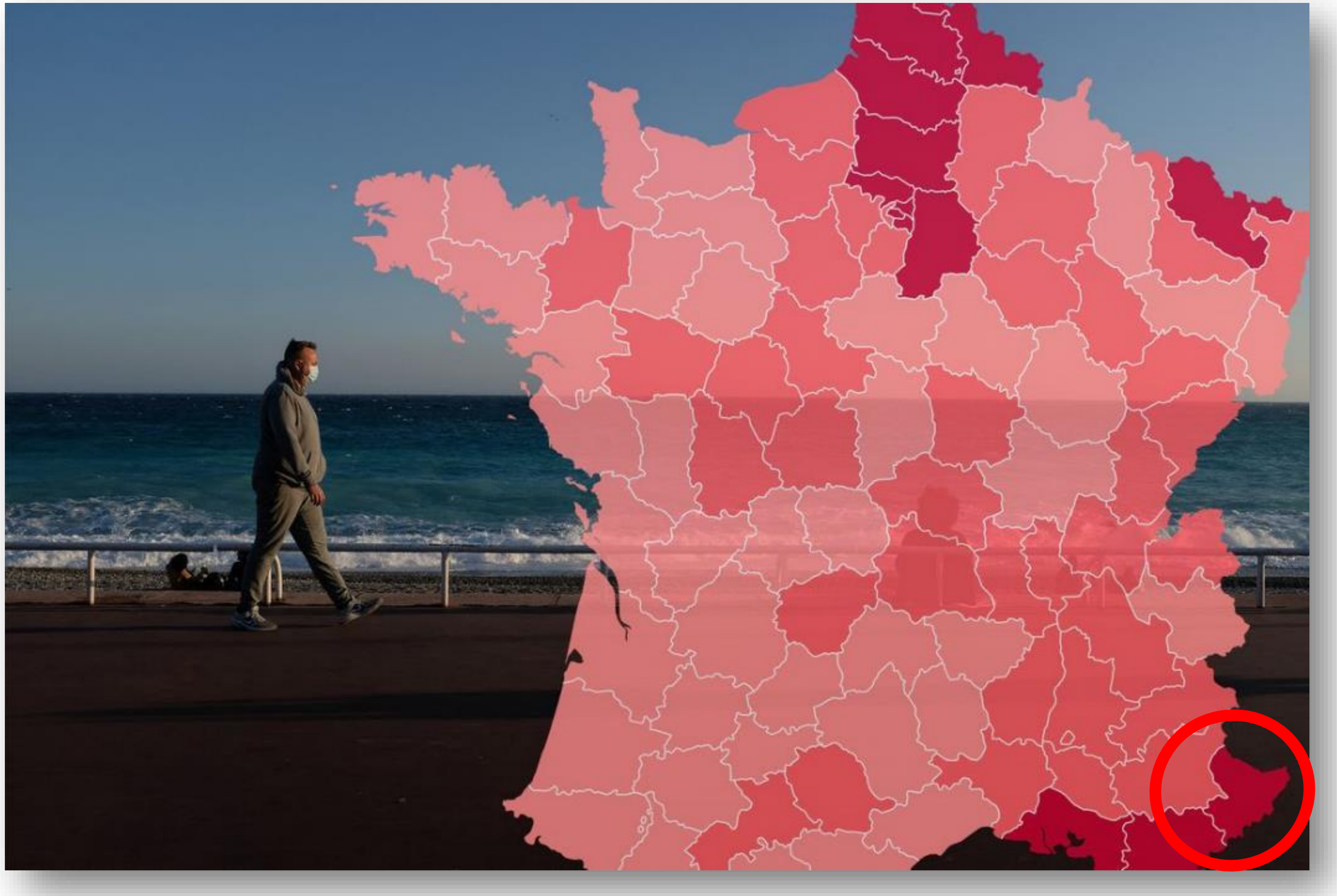
Total Test Results in US
421 219 495

58 202 413 tests	California US
49 426 695 tests	New York US
22 537 890 tests	Texas US
22 120 098 tests	Florida US
21 920 359 tests	Illinois US
20 599 779 tests	Massachusetts US
13 217 769 tests	New Jersey US
12 617 144 tests	Pennsylvania US

US Test Res...

Walk of the SARS CoV 2 on the « promenade des Anglais »

Higher incidence of the virus in France (March 2021)



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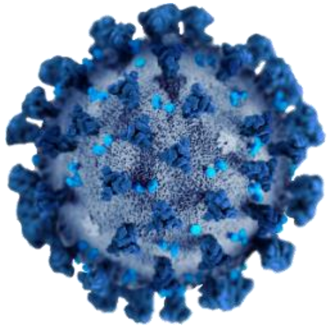

Louis Pasteur Hospital, University of Nice, France

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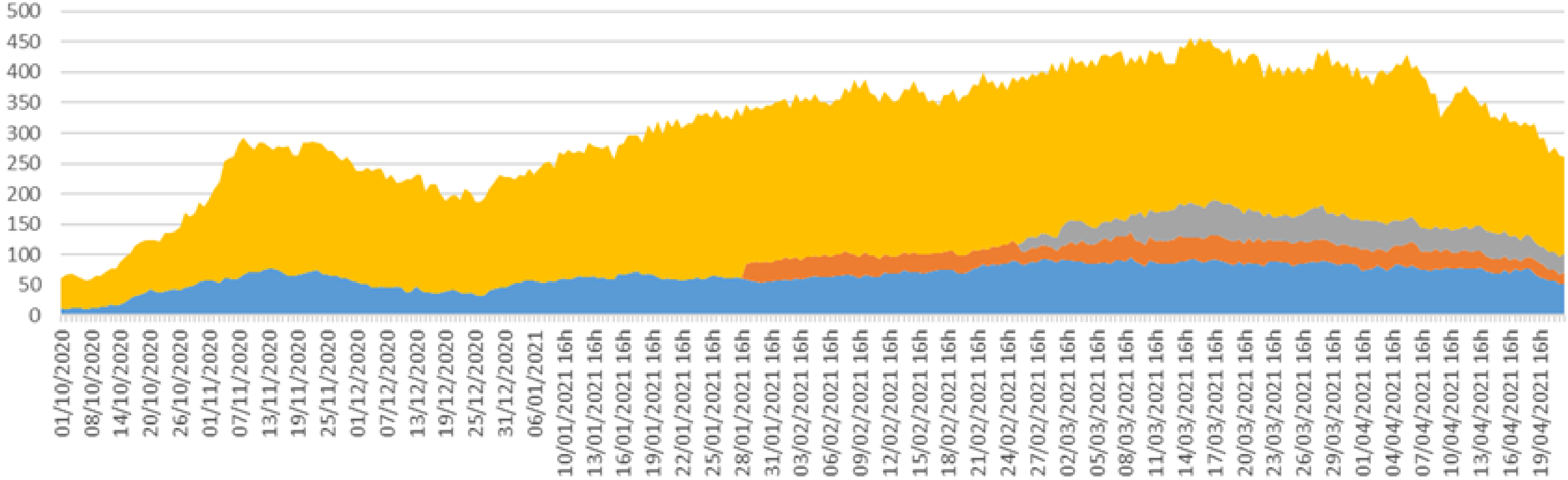
Evolution of COVID-19 cases in Nice, France

October 1, 2020 to April 19, 2021



🟨 Hospitalisation
▨ Primary schools
🟦 Intensive care unit
🟧 Residential homes

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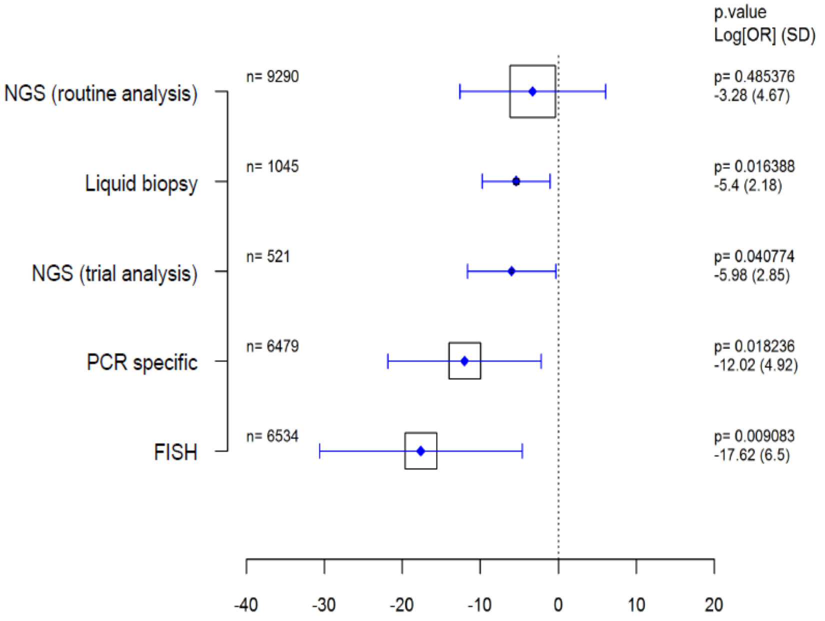
GOOD SCIENCE
BETTER MEDICINE
BEST PRACTICE

CANCER
HORIZONS

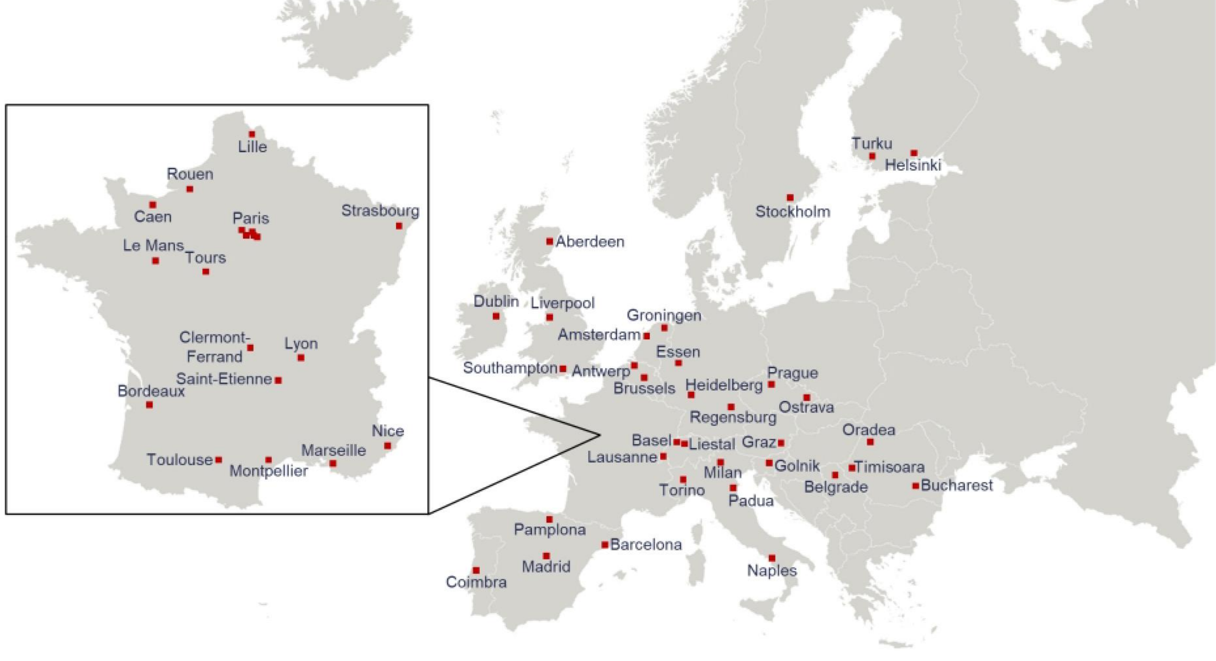
ORIGINAL RESEARCH

Clinical and molecular practice of European thoracic pathology laboratories during the COVID-19 pandemic. The past and the near future

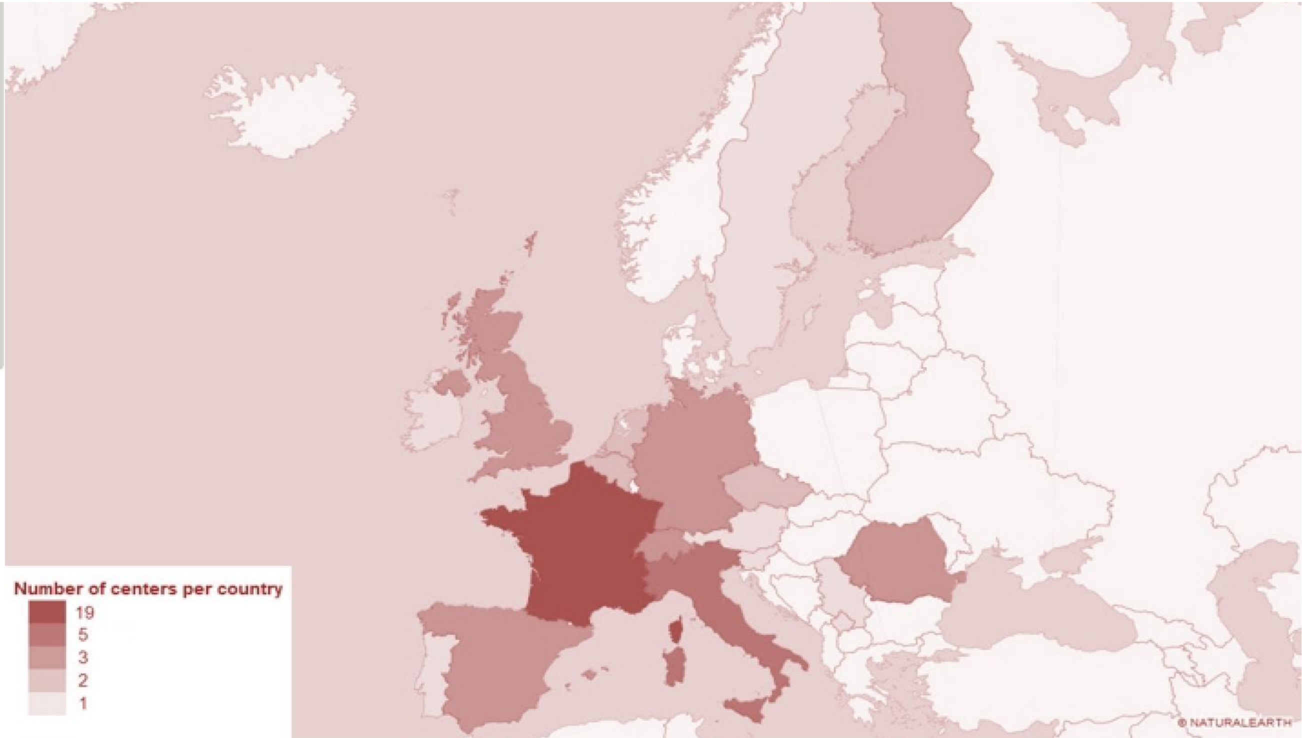
P. Hofman[1†*], M. Ilié[1†], E. Chamorey[2†], P. Brest[3†], R. Schiappa[2], V. Nakache[1], M. Antoine[4], M. Barberis[5], H. Begueret[6], F. Bibeau[7], C. Bonnetaud[1], P. Boström[8], P. Brousset[9], L. Bubendorf[10], L. Carvalho[11], G. Cathomas[12], A. Cazes[13], L. Chalabreysse[14], M.-P. Chenard[15], M.-C. Copin[16], J.-F. Côté[17], D. Damotte[18], L. de Leval[19], P. Delongova[20], V. Thomas de Montpreville[21], A. de Muret[22], A. Dema[23], W. Dietmaier[24], M. Evert[24], A. Fabre[25], F. Forest[26], A. Foulet[27], S. Garcia[28], M. Garcia-Martos[29], L. Gibault[30], G. Gorkiewicz[31], D. Jonigk[32], J. Gosney[33], A. Hofman[1], I. Kern[34], K. Kerr[35], M. Kossai[36], M. Kriegsmann[37], S. Lassalle[1], E. Long-Mira[1], A. Lupo[18], A. Mamilos[24], R. Matěj[38], J. Meilleroux[9], C. Ortiz-Villalón[39], L. Panico[40], A. Panizo[41], M. Papotti[42], P. Pauwels[43], G. Pelosi[44], F. Penault-Llorca[36], O. Pop[45], N. Poté[13], S. R. Y. Cajal[46], J.-C. Sabourin[47], I. Salmon[48], M. Sajin[49], S. Savic-Prince[10], H.-U. Schildhaus[50], P. Schirmacher[37], I. Serre[51], E. Shaw[52], D. Sizaret[22], A. Stenzinger[37], J. Stojsic[53], E. Thunnissen[54], W. Timens[55], G. Troncone[56], C. Werlein[32], H. Wolff[57], J.-P. Berthet[58], J. Benzaquen[59], C.-H. Marquette[59], V. Hofman[1†] & F. Calabrese[60†]



Molecular Biology activity from March to May 2019 and from March to May 2020

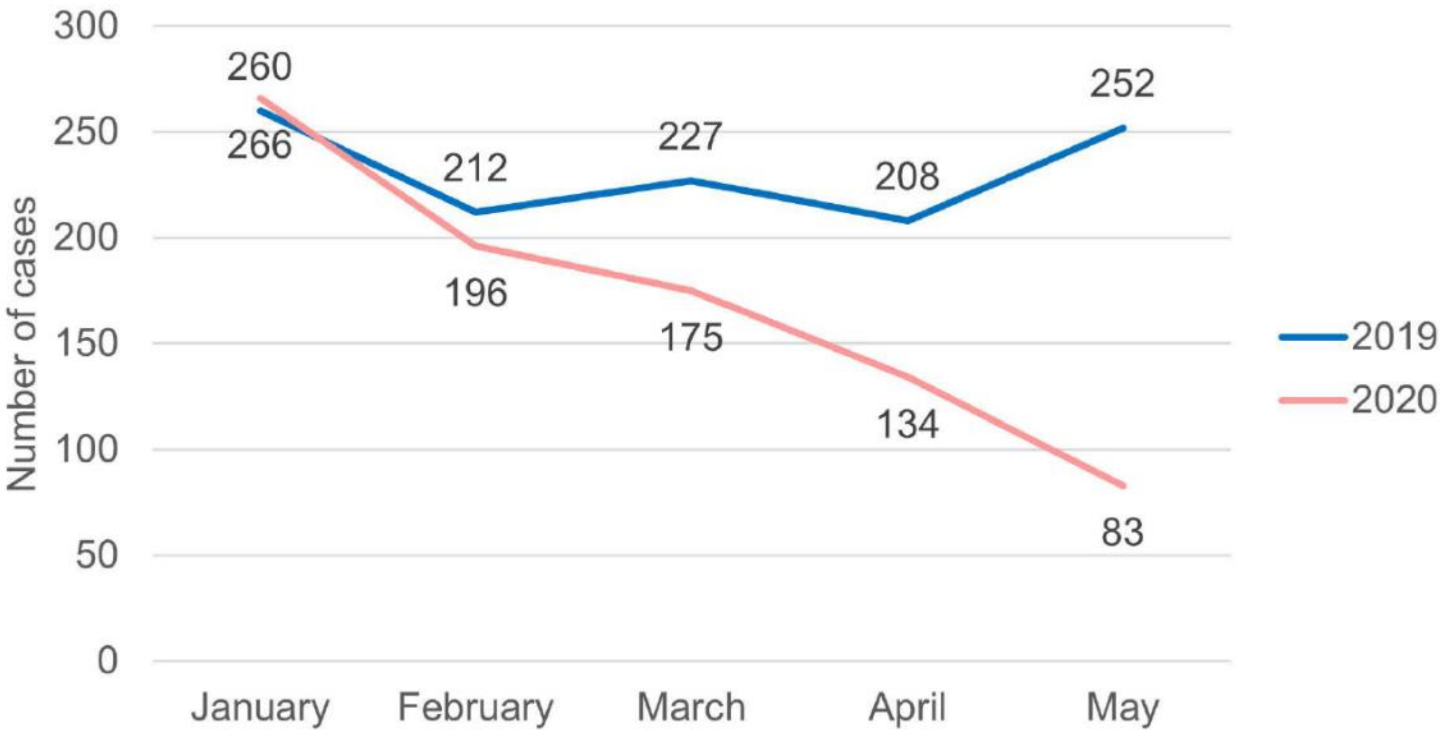


Pathology laboratories participating to the survey



Activity of thoracic cancer surgeries in the South Region, France

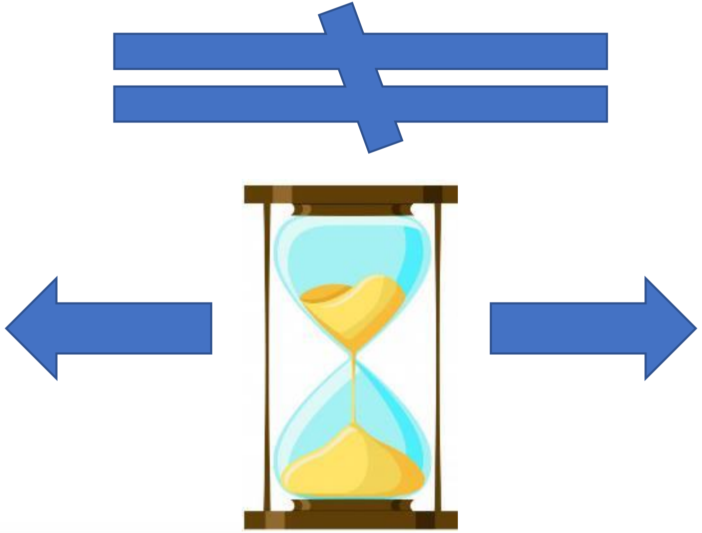
Between 2019 and 2020



ORIGINAL RESEARCH

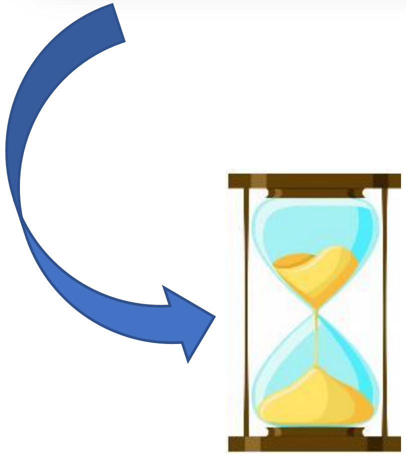
Clinical and molecular practice of European thoracic pathology laboratories during the COVID-19 pandemic. The past and the near future

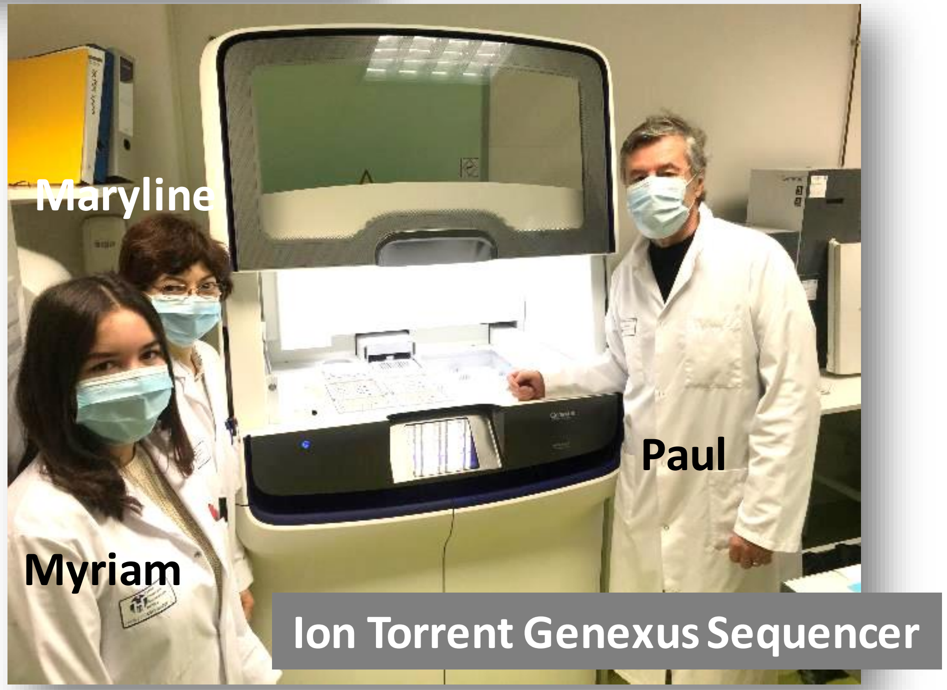
Nice (LPCE)
molecular pathology platforms



Chef & GeneStudio NGS

Targeted sequencing
Idylla (Biocartis)





Ion Torrent Genexus Sequencer

Targeted sequencing
Cobas (Roche)

Specimen to report in a single day with only two user touchpoints

Ion Torrent Genexus System

- Lysate from FFPE tissue
- Plasma
- Whole blood
- Peripheral blood lymphocyte (PBL)
- Lysate from fresh-frozen tissue
- Lysate form bone marrow

Genexus Software

Nucleic acid purification and quantitation*

Ion Torrent™ Genexus™

Purification System (Available 2021)

2 hour turnaround time

12 FFPE (DNA and RNA)

6 Plasma

Library preparation to variant interpretation

Report*

Ion Torrent™ Genexus™

Integrated Sequencer (November 2019)

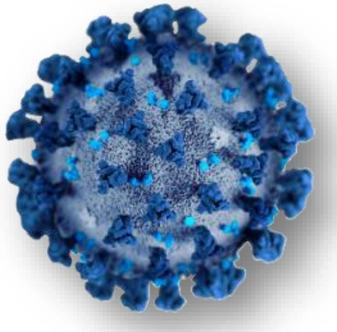
Ion Torrent™ GX5™ Chip:

12–15M reads/lane

14 hours for a single-lane run

(approx. 24 to 30 hours for full chip)

Up to 32 Samples per run



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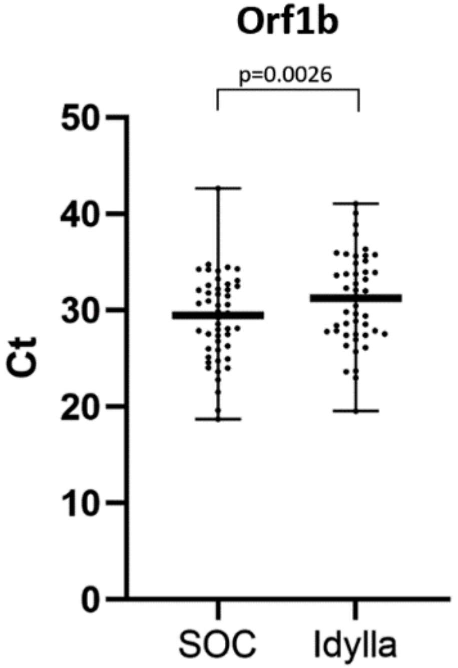
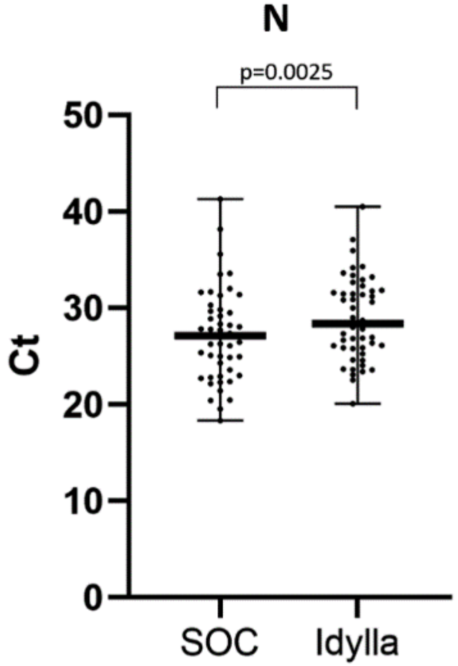
Setting up a collection of SARS-CoV-2 positive NSP samples and storage in an accredited biobank (BB-0033-00025)

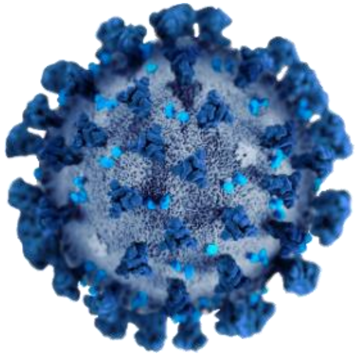
First period



SOC = 29.1 ± 4.55 [range 18.7 - 42.6]
Idylla = 31.0 ± 4.66 [range 19.5 - 41.0]



SOC = 27.2 ± 4.7 [range 18.3 – 41.3]
Idylla = 28.9 ± 4.3 [range 20.1 – 40.5]



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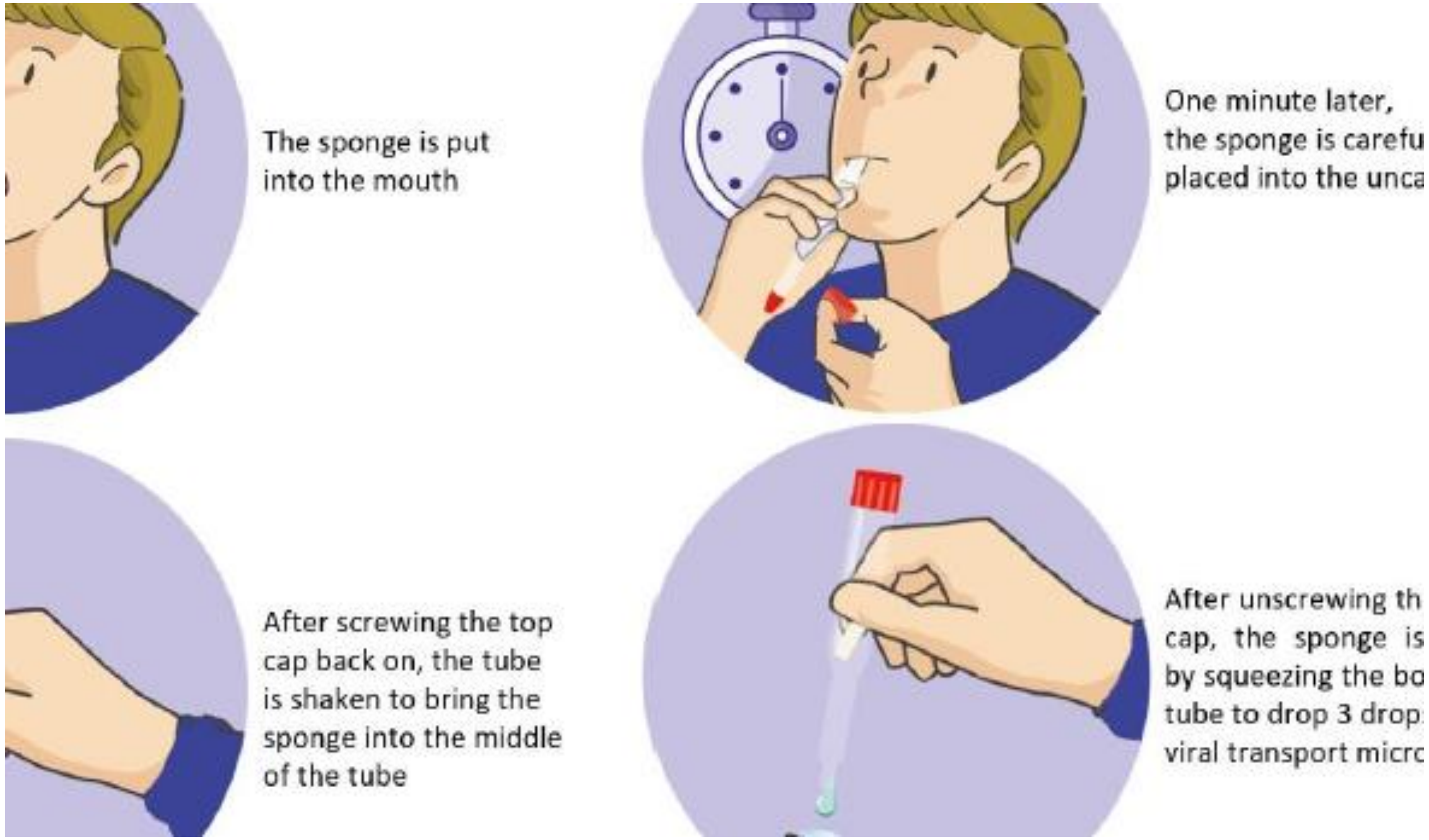
Hofman et al, Ann Transl Med 2021, in press

Setting up a collection of SARS-CoV-2 positive oral samples and storage in an accredited biobank (BB-0033-00025)

Second period

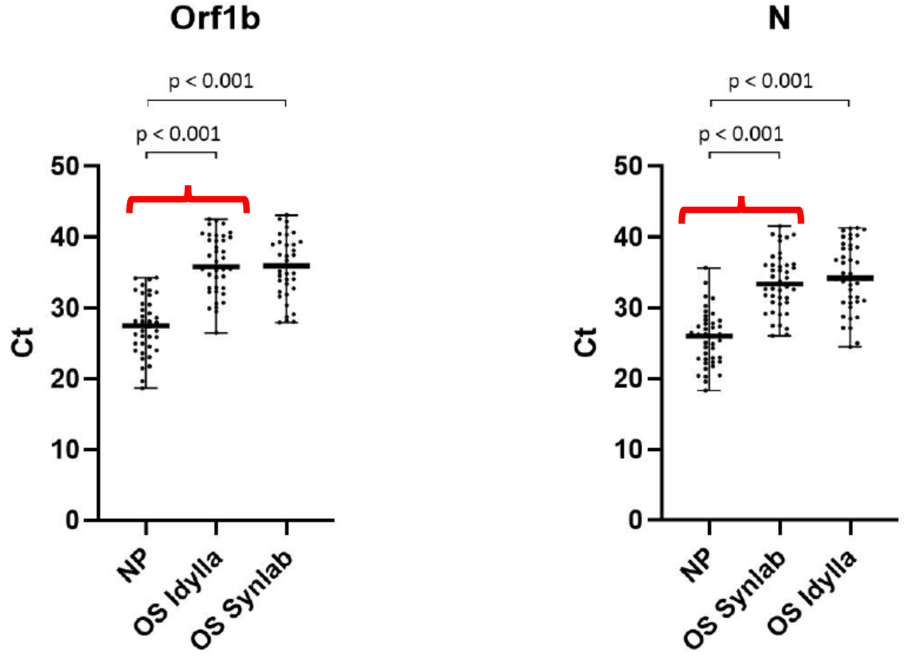

The sponge is put into the mouth

One minute later, the sponge is carefu placed into the unca

After screwing the top cap back on, the tube is shaken to bring the sponge into the middle of the tube

After unscrewing th cap, the sponge is by squeezing the bo tube to drop 3 drop viral transport micro

All-in-on saliva sampling technique with a sponge.
Usefulness of a double-capped plastic



Viral load of NasoPharyngeal and oral samples assessed by the Ct for the *Orf1b* and *N* genes

Manuscript submitted, 2021

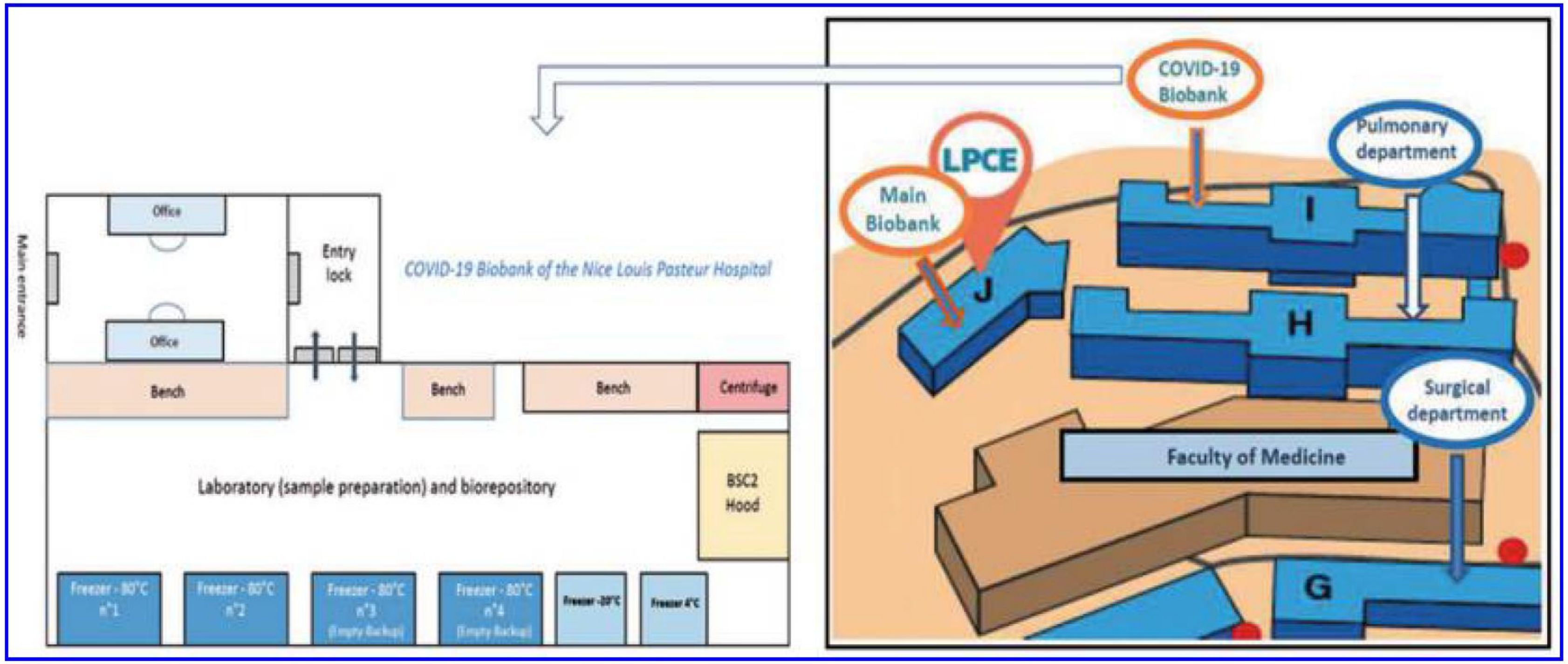
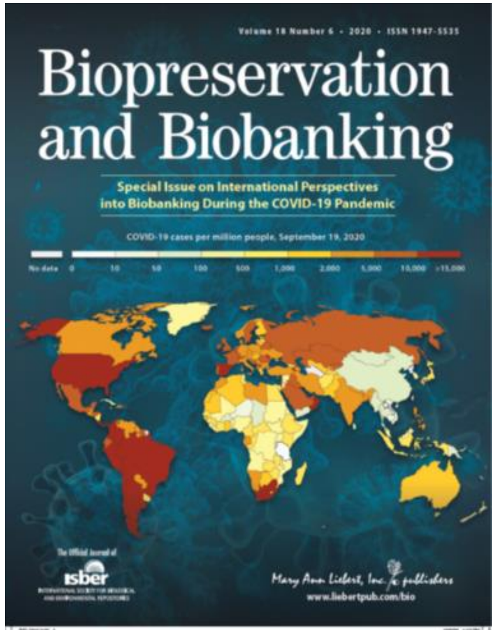


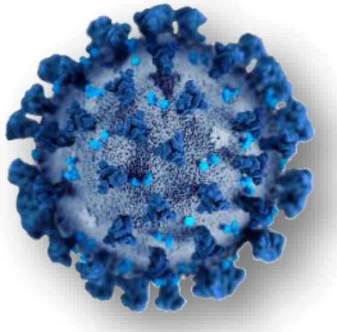
FIG. 1. Facilities of the COVID-19 Nice Louis Pasteur Hospital Biobank. LPCE, Laboratory of Clinical and Experimental Pathology.



BIOPRESERVATION AND BIOBANKING
Volume 00, Number 00, 2020
© Mary Ann Liebert, Inc.
DOI: 10.1089/bio.2020.0055

Establishment of a Collection of Blood-Derived Products from COVID-19 Patients for Translational Research: Experience of the LPCE Biobank (Nice, France)

Virginie Tanga,[1,2] Sylvie Leroy,[3] Julien Fayada,[1] Marame Hamila,[1] Maryline Allegra,[1]
Zeineb Messaoudi,[1] Christelle Bonnetaud,[1] Virgine Lespinet,[1] Olivier Bordone,[1]
Kevin Washetine,[1] Jennifer Griffonnet,[3] Charlotte Maniel,[3] Lorène Philibert,[3] Eric Selva,[1]
Jonathan Benzaquen,[3] Marius Ilie,[1,2] Elodie Long,[1,2] Sandra Lassalle,[1,2] Elisabeth Lantéri,[1,2]
Charles-Hugo Marquette,[2,3] Véronique Hofman,[1,2] and Paul Hofman[1,2]



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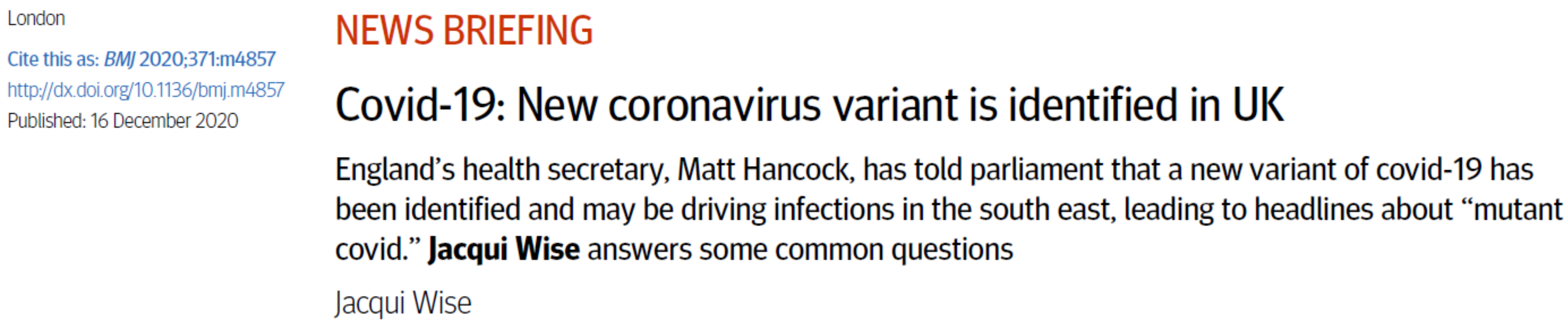


London

Cite this as: *BMJ* 2020;371:m4857

http://dx.doi.org/10.1136/bmj.m4857

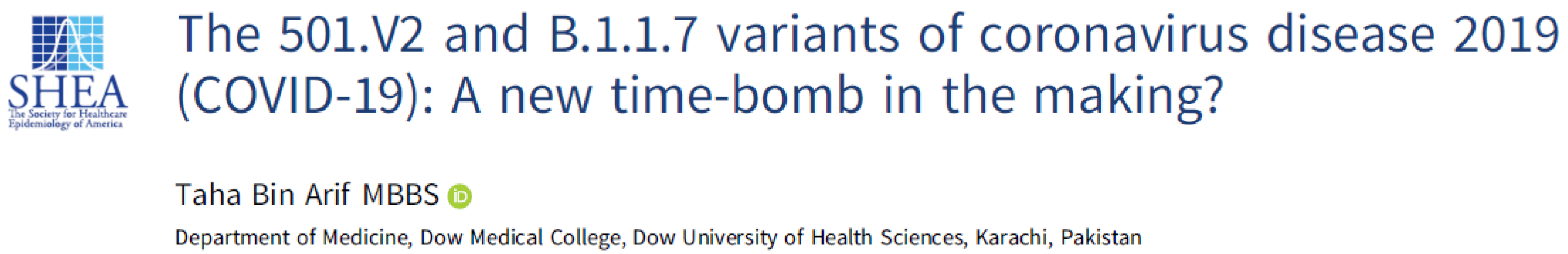
Published: 16 December 2020

NEWS BRIEFING

Covid-19: New coronavirus variant is identified in UK

England's health secretary, Matt Hancock, has told parliament that a new variant of covid-19 has been identified and may be driving infections in the south east, leading to headlines about "mutant covid." **Jacqui Wise** answers some common questions

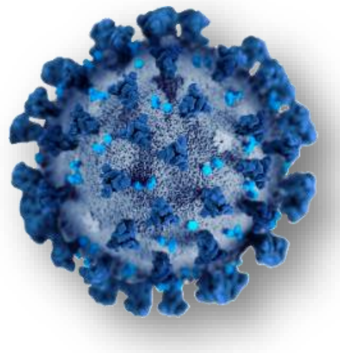
Jacqui Wise



The 501.V2 and B.1.1.7 variants of coronavirus disease 2019 (COVID-19): A new time-bomb in the making?

Taha Bin Arif MBBS

Department of Medicine, Dow Medical College, Dow University of Health Sciences, Karachi, Pakistan



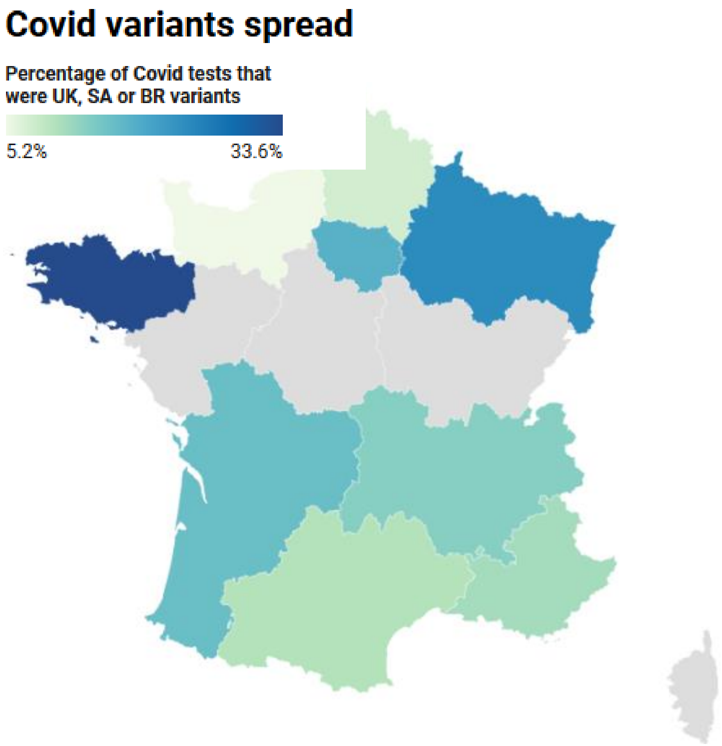
Could it be that the B.1.1.7 lineage is more deadly?

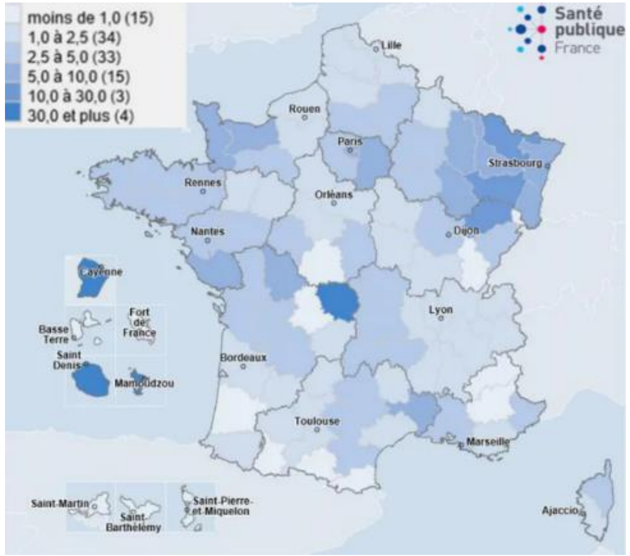
Chia Siang Kow[1] and Syed Shahzad Hasan[2,3]

[1]School of Postgraduate Studies, International Medical University, Kuala Lumpur, Malaysia, [2]School of Applied Sciences, University of Huddersfield, Huddersfield, United Kingdom and [3]School of Biomedical Sciences & Pharmacy, University of Newcastle, Callaghan, Australia

SARS COV 2 variants of concerns present in France

- B.1.17 (**UK**): 82% of variants in France April 21, 2021
- 20H/501Y.V2 (**South of Africa**) and 20J/501Y.V3 (**Brazil**): 4.2 % of variants in France April 21, 2021
- B.1.616 (ou 20C/655Y) (**Britain, France**): Not detected using SARS-CoV-2 RT-PCR from NSP swabs


Covid variants spread



April 21, 2021, % of 20H/501Y.V2 and 20J/501Y.V3 variants among SARS-CoV-2 positive tests

B.1.617 (India)

April 29, 2021


Covid-19 dans le monde : le variant indien détecté en Belgique, chez des étudiants passés par Roissy

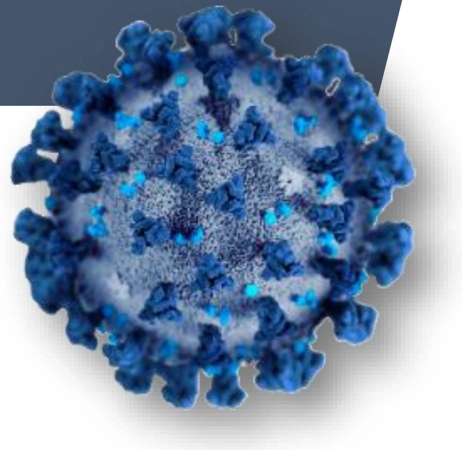
Vingt étudiants indiens ont été testés positifs à ce mutant du SARS-CoV-2. Ils ont été placés en quarantaine dans les villes flamandes d'Alost et de Louvain, où ils étaient arrivés mi-avril.

Le Monde ·

Key Questions around the new Coronavirus Strains

- How can we detect new strains?
- What is the origin of the new strain and how prevalent is it?
- Will current molecular or antigen-based tests miss detecting this strain?
- Does this strain spread more quickly?
- **Does this strain lead to higher disease severity?**
- **What is the vaccine efficacy against the new strain?**

There are still currently many unknowns.
Ongoing research is needed to better understand virus strain evolution and spread.



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Key questions on the new Coronavirus strains

- How can we detect
- What is the origi
  prevalent is it?
- Will current m
  detecting this
- Does this str
- **Does this strai**
- **What is the vaccine effica**

There are still currently many
nknowns.
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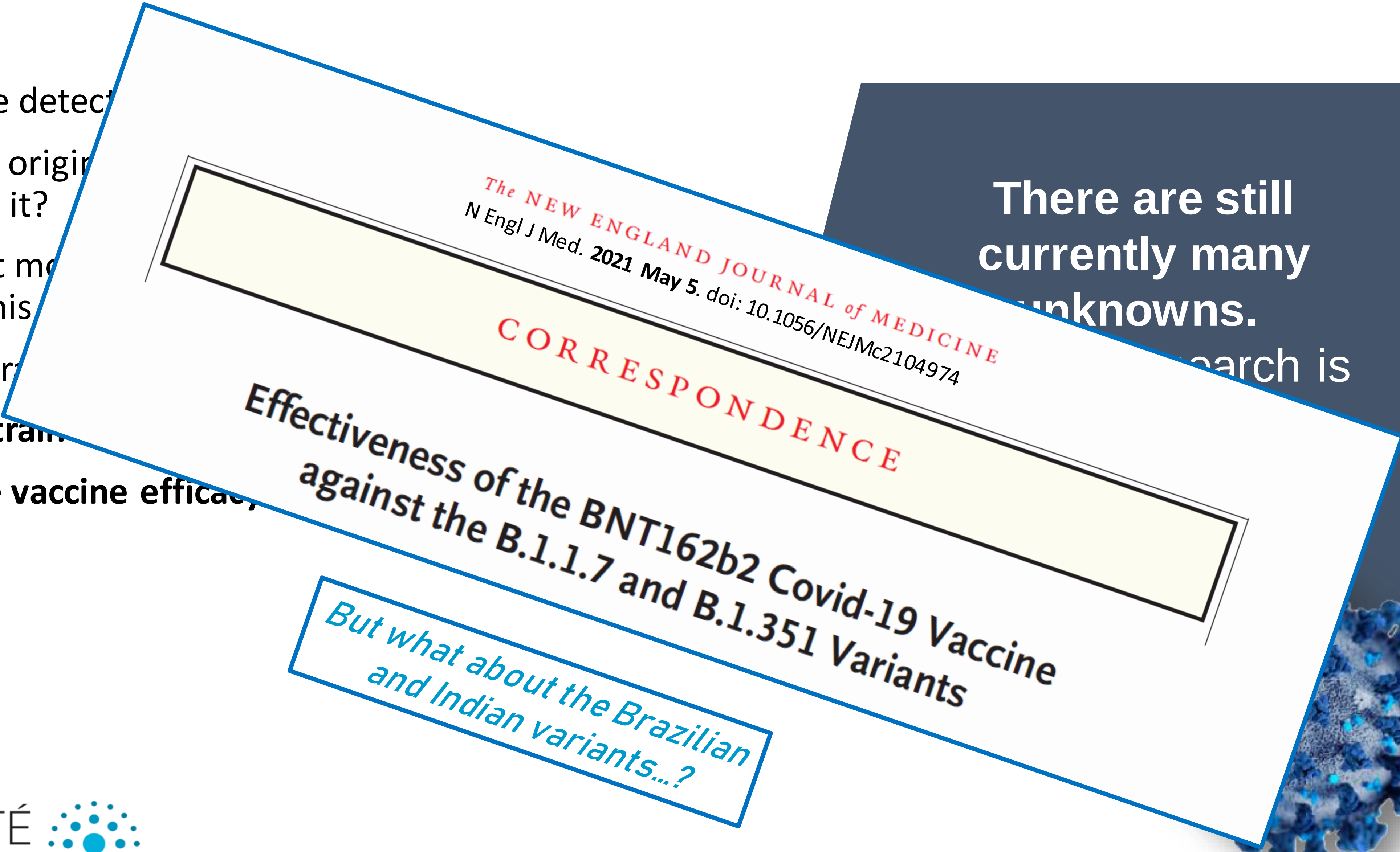
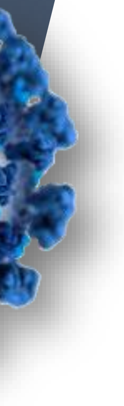


The NEW ENGLAND JOURNAL of MEDICINE
N Engl J Med. **2021 May 5**. doi: 10.1056/NEJMc2104974

CORRESPONDENCE

Effectiveness of the BNT162b2 Covid-19 Vaccine against the B.1.1.7 and B.1.351 Variants

But what about the Brazilian and Indian variants…?



Retrospective analysis

Starting March 2021

145 nasopharyngeal swabs samples and 145 matched saliva samples positives for RT-PCR SARS-CoV-2

Individuals tested from *April 2020 to January 2021*

Samples collected and stored at -80°C in the Biobank 0033-00025 University Côte d'Azur (www.biobank-cotedazur.fr)

From RT-PCR to NGS assessment of SARS-CoV-2

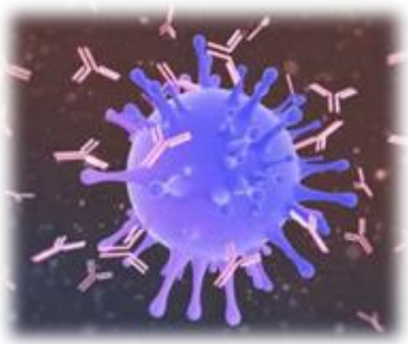
Prospective analysis

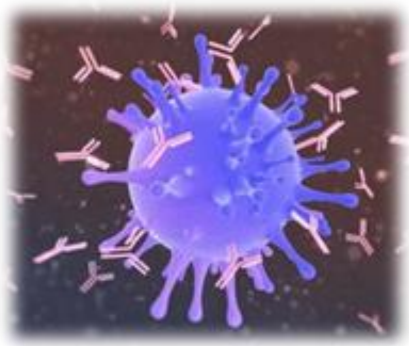
Starting April 2021

Systematic screening of NSP swabs positive for RT-PCR SARS-CoV-2 but without any variants identification using the commercially multiplex test

Screening of NSP swabs with clinically suspected COVID-19 but negative for RT-PCR SARS-CoV-2

Screening of NSP swabs in some specified clusters





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Projects started in 2021 at the LPCE (Nice, France)

Ion AmpliSeq SARS-CoV-2 Research Panel

One assay surveying complete SARS-CoV-2 genome for epidemiological investigation

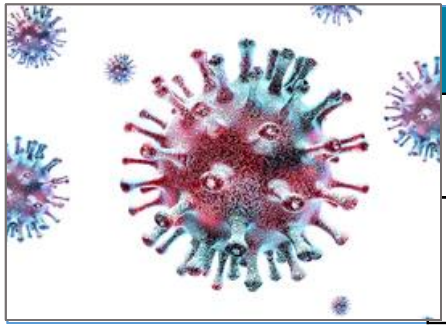
1-10 ng total RNA

1 ng cDNA input per pool

Nucleic acid sample input

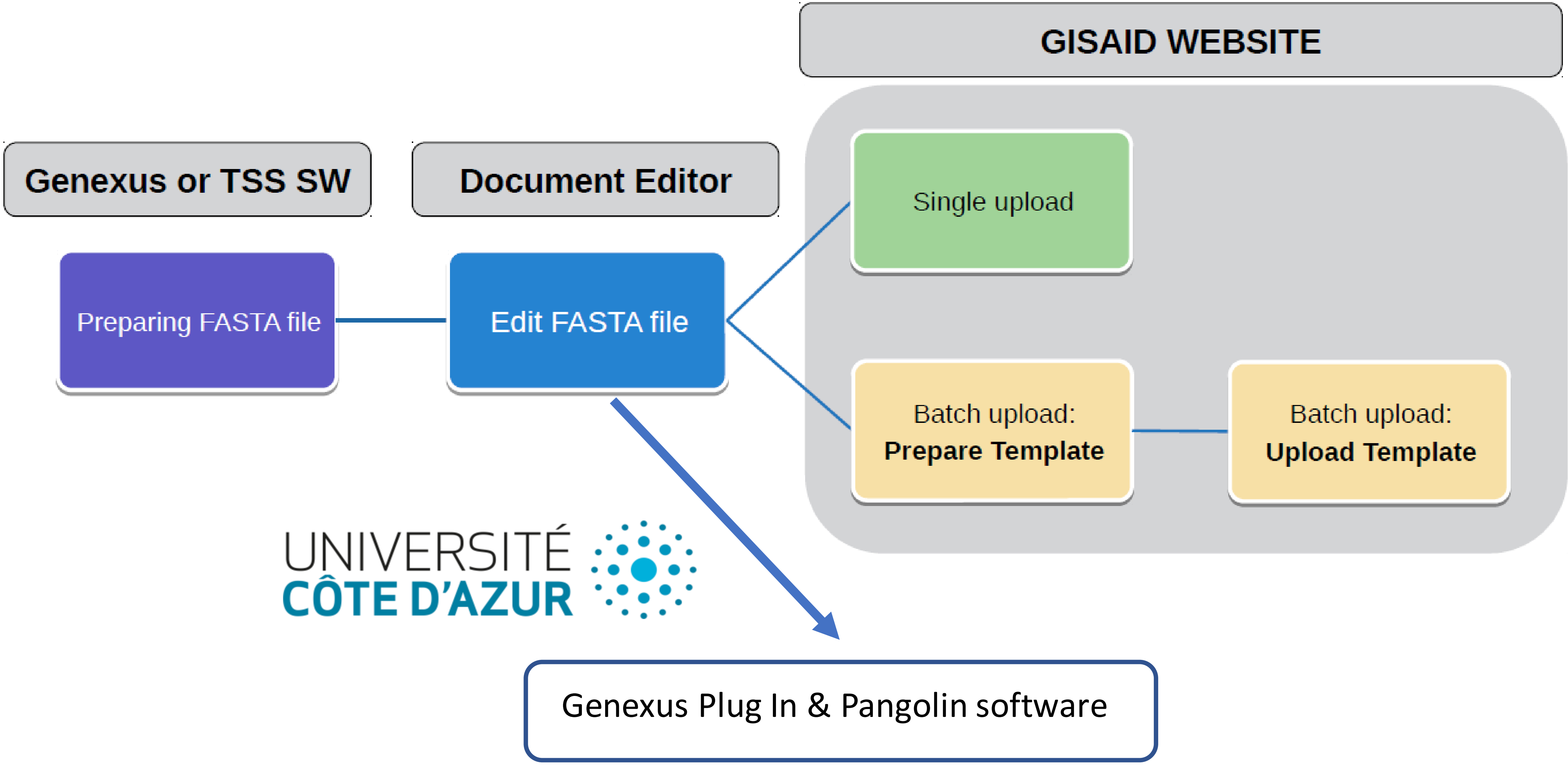
Automate cDNA synthesis library prep, template prep, sequencing and variant analysis

- **>99% coverage of SARS-CoV-2 genome (~30kb)**
- **World leading amplification technology: AmpliSeq™**
- Amplicon length range: 125-275 bp
- 237 amplicons specific to SARS-CoV-2 + 5 human expression controls
- All potential serotypes covered
- 2250 manual-preped, 600 Chef-automated libraries or 240 Genexus-automated libraries



Pool	No. Total Amplicons
1	**247 (242 unique)***
2	

Data analyses for SARS COV 2 variants



GISAID WEBSITE

Genexus or TSS SW

Document Editor

Preparing FASTA file

Edit FASTA file

Single upload

Batch upload:
Prepare Template

Batch upload:
Upload Template

Genexus Plug In & Pangolin software

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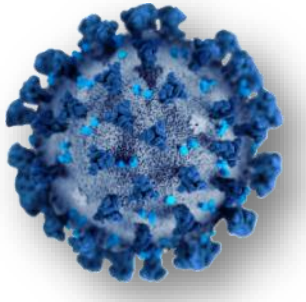
Identified variants in a retrospective analysis

Ongoing study conducted on the Ion Torrent Genexus system
total, 102 samples

Variants	
B.1.160 (European)	64
B.1.1.77 (UK)	17
B.1.1.7 (UK)	**8**
B.1.241 (European)	3
B.23 (UK): 5 (4%)	2
B.1.1.2.41 (European)	1
B.1.1.2.21 (European)	1
B.1.596 (USA)	1
B.1.160.8 (Denmark)	1
A.19 (Burkina Faso)	1
B.1.411 (Sri Lanka)	1



Variant classification

- **VOC (variants of concern)**
  - B.1.1.7 (UK)
  - B.1.351(South Africa)
  - B.1.1.28.1 (Brazil)
  - B.1.617 (India)
- **VOI (variants under investigation)**
- **VUM (variant under monitoring)**

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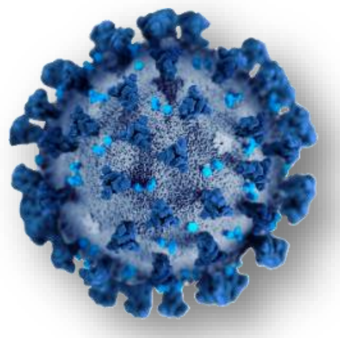
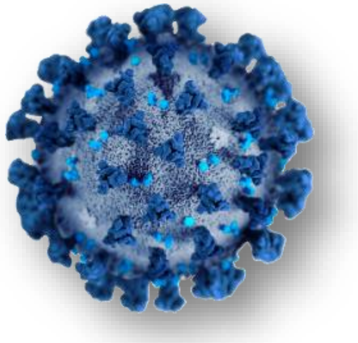
Comparative analyses obtained from matched nasopharyngeal swabs (NSP) and saliva samples

Samples	NSP 	Saliva 
1	B.1.160	B.1.160
2	B.1.1.7	B.1.1.7
3	B.1.160	B.1.160
4	B.1.1.2.41	B.1.1.2.41
5	A.19	A.19
6	B.1.411	B.1.411
7	B.1.596	B.1.596
8	B.1.177	B.1.177
9	B.1.160	B.1.160
10	B.1.160	failed
11	B.1.160	B.1.160
12	B.1.1.7	B.1.1.7
13	B.1.160	B.1.160
14	B.1.160	B.1.160



GENEXUS: More advantages, less constraints during the COVID-19 crisis

- Less contact between the health care workers during the manipulation (better biosafety)
- Easy to use
- Easy to analyse data
- Short turnaround time (24h)
- Workflow automation
- Flexible (integration of different workflows for genomic assessment of solid tumors too)
- High number of analyses (until 112 samples per week)
- High **sensitivity** of detection (able to detect variants even with a low viral load/high Ct)
- Panel design improvement Ion AmpliSeq™ SARS-CoV-2 Insight Research Panel – just launched



The COVID-19 crisis in Nice, France, and the impact in a pathology lab

SARS-CoV-2 diagnosis set-up in a pathology lab: experience of the LPCE

Current development using the NGS approach for SARS-CoV-2 assessment

Perspective

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www.thelancet.com/infection Vol 21 January 2021

Genomic evidence for reinfection with SARS-CoV-2: a case study

Richard L Tillett, Joel R Sevinsky, Paul D Hartley, Heather Kerwin, Natalie Crawford, Andrew Gorzalski, Chris Laverdure, Subhash C Verma, Cyprian C Rossetto, David Jackson, Megan J Farrell, Stephanie Van Hooser, Mark Pandori

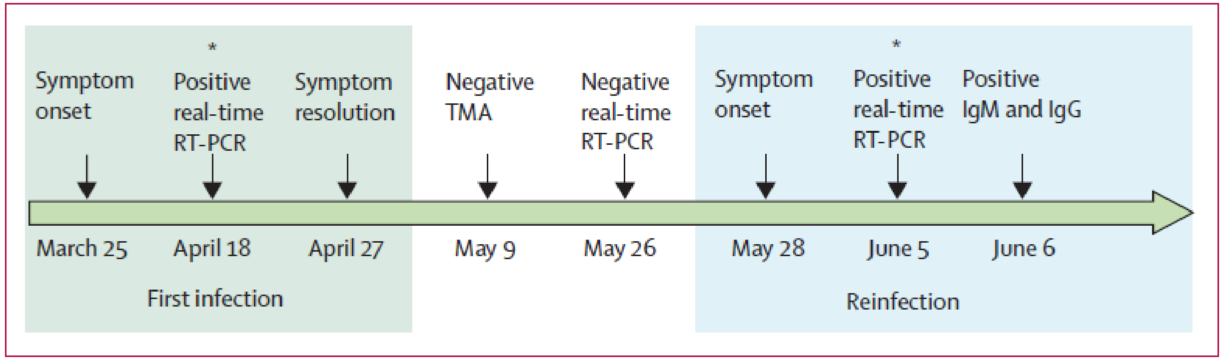

Figure 1: **Timeline of symptom onset, molecular diagnosis, and sequencing of specimens**
TMA=transcription-mediated amplification. *Sequenced specimens.

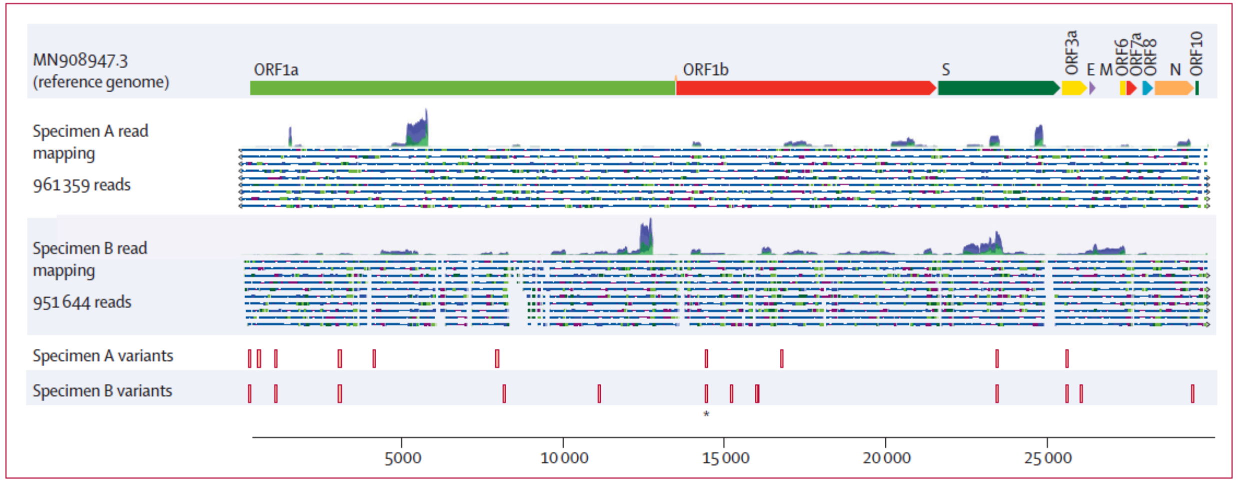

Figure 2: **Variant mapping of specimens A and B against the reference genome**
ORF1a and ORF1b encode replicase proteins. The other ORFs encode assembly proteins. ORF=open reading frame. S=spike. E=envelope. M=membrane. N=nucleocapsid. *Identifies variant 14 407 in specimen A and variants 14 407 and 14 408 in specimen B.


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MINIREVIEW

The Importance and Challenges of Identifying SARS-CoV-2 Reinfections

Ahmed Babiker,[a,b] Charles E. Marvil,[b] Jesse J. Waggoner,[a] Matthew H. Collins,[a] Anne Piantadosi[a,b]

Emerging variants
to be permanently detected

The world this week

News in focus

Nature | Vol 589 | 14 January 2021 | 177

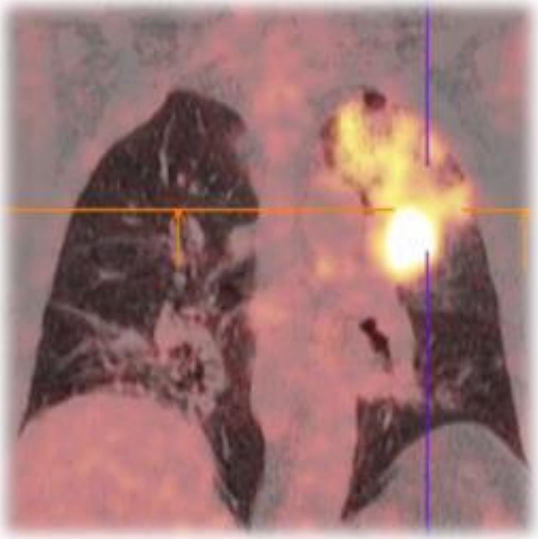
ospital worker monitors a person with COVID-19 in South Africa, where a new variant of SARS-CoV-2 has been identified.

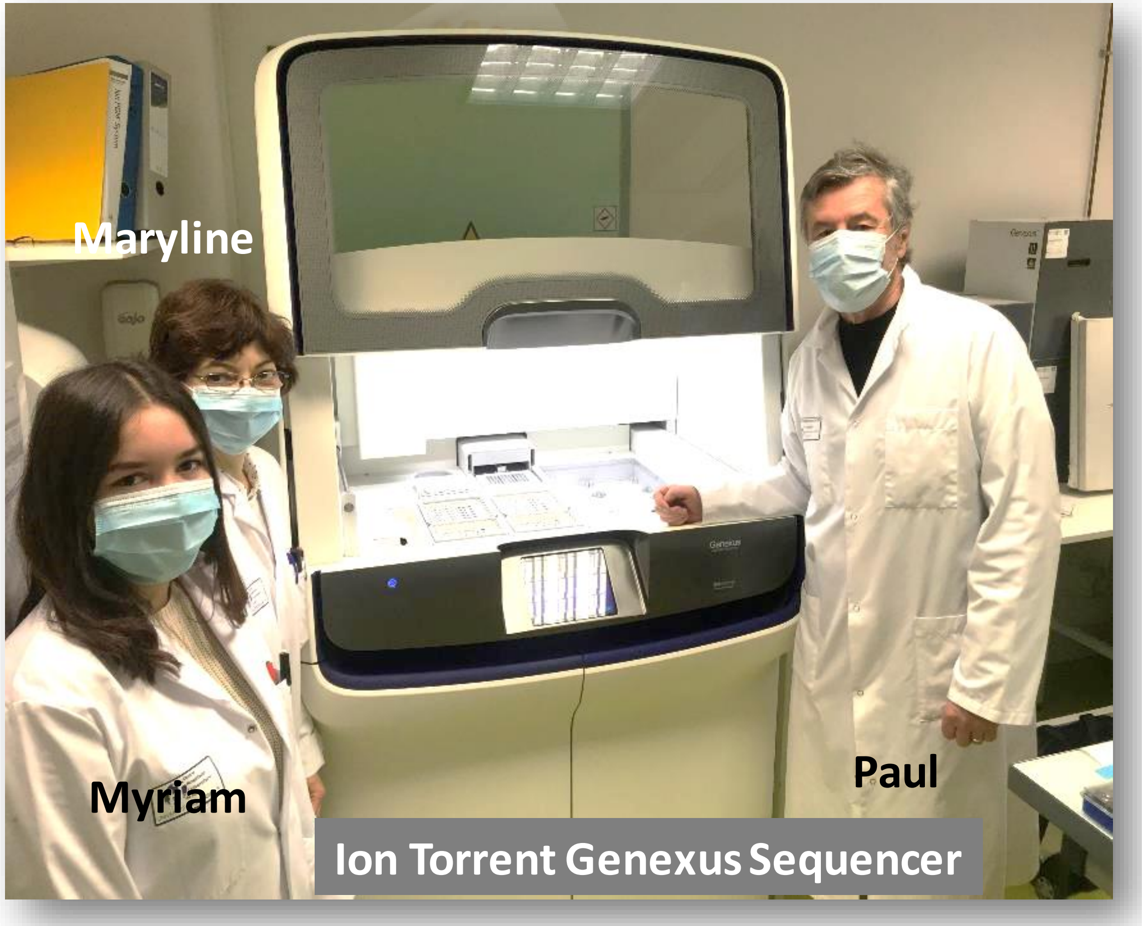
COULD NEW COVID VARIANTS UNDERMINE VACCINES? LABS SCRAMBLE TO FIND OUT

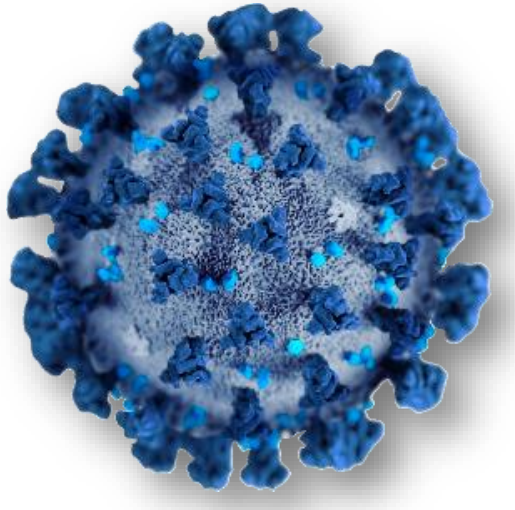
Researchers race to determine why lineages identified in Britain and South Africa spread so quickly, and whether vaccines will be less effective against them.

INDIA'S DOUBLE MUTANT VARIANT 'CONCERN'

Integrating the activities in the same platform







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