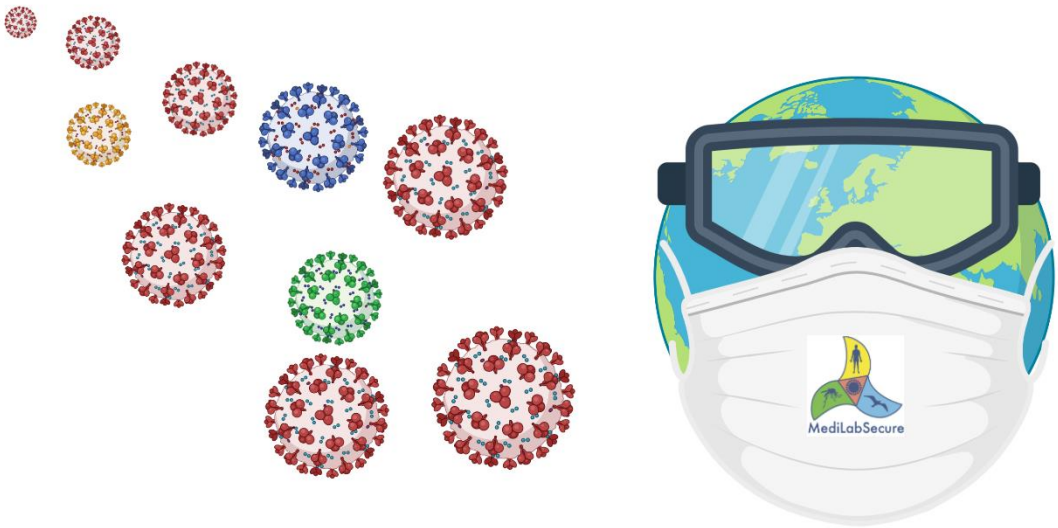
MediLabSecure human virology – Newsletter n°7 – January 2021

HUMAN VIROLOGY NETWORK of MEDILABSECURE

Newsletter #7

The newsletter from the human virology group of MediLabSecure



Newsletter special COVID-19 variants

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A new phase of the COVID-19 pandemic started with the multiplication of viral variants with problematic characteristics such as increased dispersion, immune evasion, laboratory identification technics tempered and need for more complex and quick identification of these variants worldwide. Here is a quick overview of what we can do at the laboratory level and within MediLabSecure.

- **SARS-CoV-2 variants:**

➢ Current situation

On 14 December 2020, authorities of the United Kingdom reported to WHO a variant referred to by the United Kingdom as SARS-CoV-2 **VOC 202012/01** (Variant of Concern, year 2020, month 12, variant 01, also known as lineage **B.1.1.7** or 20I/501Y.V1 (formerly 20B/501Y.V1)). This variant contains 23 nucleotide substitutions and is not phylogenetically related to the SARS-CoV-2 virus circulating in the United Kingdom at the time the variant was detected. Preliminary epidemiologic, modelling, phylogenetic and clinical findings suggest that SARS-CoV-2 VOC 202012/01 has increased transmissibility. However, preliminary analyses also indicate that there is no change in disease severity (**!** Caution, recent announcement by UK prime minister suggests the variant might be more severe**!**), or occurrence of reinfection between variant cases compared to other SARS-CoV-2 viruses circulating in the United Kingdom. As of 13 January, VOC-202012/01 variant has been reported in 50 countries/territories/areas worldwide.

(adapted from WHO website, 31/12/2020).

Rambaut A, Loman N, Pybus OG, et al. Preliminary genomic characterisation of an emergent SARS-CoV-2 lineage in the UK defined by a novel set of spike mutations. Virological, December, 2020. https://virological.org/t/preliminary-genomic-characterisation-of-anemergent-sars-cov-2-lineage-in-the-uk-defined-by-a-novel-set-of-spikemutations/563

On 18 December, national authorities in South Africa announced the detection of a new variant of SARS-CoV-2 that is rapidly spreading in three provinces of South Africa. South Africa has named this variant **501Y.V2** (also known as **501.V2**, 20H/501Y.V2 (formerly 20C/501Y.V2), or lineage **B.1.351**), because of a N501Y mutation. While SARS-CoV-2 VOC 202012/01 from the UK also has the N501Y mutation, phylogenetic analysis has shown that 501Y.V2 from South Africa are different virus variants. In the week beginning 16 November, routine sequencing by South African health authorities found that this new SARS-CoV-2 variant has largely replaced other SARS-CoV-2 viruses circulating in the Eastern Cape, Western Cape, and KwaZulu-Natal provinces. This variant is thought to spread faster and might have a capacity to escape immunity in immunized people. As of 30 December, the 501Y.V2 variant from South Africa has been reported from four other countries to date worldwide.

(adapted from WHO website, 31/12/2020)

Tegally H, Wilkinson E, Giovanetti M, et al. Emergence and rapid spread of a new severe acute respiratory syndrome-related coronavirus 2 (SARS-CoV-2) lineage with multiple spike mutations in South Africa. MedRxiv 2020; published online Dec 22. https://doi.org/10.1101/2020.12.21.20248640 (preprint).

Lineage P.1 was initially detected in Tokyo on 6 January 2021 by the National Institute of Infectious Diseases (NIID) in four people who arrived from the Amazonas state. This variant of SARS-CoV-2 is a descendant of B.1.1.28 and has been named P.1 lineage, it has 17 unique amino acid changes, 10 of which in its spike protein, including N501Y and E484K. This variant is thought to spread faster and might have a capacity to escape immunity in previously infected people. The new lineage is currently responsible for a large outbreak in Manaus, a city that was

said to have ~75% of its population immunized against COVID-9 after the first wave last summer. (adapted from Wikipedia, oh no he didn't! yes, he did ;)

Faria NR, Claro IM, Candido D, et al. Genomic characterisation of an emergent SARS-CoV-2 lineage in Manaus: preliminary findings. Virological, January, 2021. https://virological.org/t/genomic-characterisation-of-anemergent-sars-cov-2-lineage-in-manaus-preliminary-findings/586.

www.thelancet.com Published online January 27, 2021 https://doi.org/10.1016/S0140-6736(21)00183-5

Other variants have been identified over the past few months such as lineage **B.1.1.207** in Nigeria (also known as **VOC-202012/01**); **Cluster 5** (AKA **ΔFVI-spike**) in the Mink farms of Denmark, that is believed to have a "moderately decreased sensitivity to neutralizing antibodies"; **lineage B.1.429** (also known as **CAL.20C**) identified in Southern California and that is spreading very fast.

WHO advises countries, where feasible, to increase routine systematic sequencing of SARS-CoV-2 viruses to better understand SARS-CoV-2 transmission and **to monitor for the emergence of variants**. Where limited sequencing capacity exists, countries are encouraged to increase capacity in collaboration with public, academic and private sequencing laboratories, and may arrange sequencing at collaborating laboratories in the COVID-19 reference laboratory network.

➢ Molecular diagnostics: qRT-PCR kits

Mutations in the sequence of the SARS-CoV-2 are of concern for the capacities of current established qRT-PCR to work correctly on all variants. For example, the situation happened in May 2020 for one commercial system (cobas® SARS-CoV-2 from Roche). However, most PCR assays in use worldwide will use multiple targets and therefore the impact of the variant on diagnostics is not anticipated to be significant.

Artesi M, et al., A Recurrent Mutation at Position 26340 of SARS-CoV-2 Is Associated with Failure of the E Gene Quantitative Reverse Transcription-PCR Utilized in a Commercial Dual-Target Diagnostic Assay. J Clin Microbiol. 2020 Sep 22;58(10):e01598-20. doi: 10.1128/JCM.01598-20. PMID: 32690547; PMCID: PMC7512182.

When sequencing solution are limited or non-existent, **qRT-PCR can be used as partial screening solution for variants**. As of today, commercial solution are slowly appearing to help laboratories.

- One of the mutations of the **VOC 202012/01** variant, the deletion at position 69/70del was found to affect the performance of the TaqPath COVID-19 Multiplex Diagnostic Solution form Thermofisher. The S gene target of the TaqPath RTPCR assay is tempered by tise deletion. This can actually be used to screen the samples for pre-identification of the UK variants.

Bal A, et al., COVID-Diagnosis HCL Study Group. Two-step strategy for the identification of SARS-CoV-2 variant of concern 202012/01 and other variants with spike deletion H69-V70,

France, August to December 2020. Euro Surveill. 2021 Jan;26(3). doi: 10.2807/1560-7917.ES.2021.26.3.2100008. PMID: 33478625.

- Our colleagues from Turkey developed an assay to pre-screen the UK, SA and Brazilian variants. Congrats and thank you for sharing!

Gulay Korukluoglu et al., 40 minutes RT-qPCR Assay for Screening Spike N501Y and HV69-70del Mutations, https://www.biorxiv.org/content/10.1101/2021.01.26.428302v1

- TibMolbiol developed a solution for the discrimination of the N501Y SNIPs (UK and SA variants) (Thank you to Jalal for the references of the kit).

VirSNiP SARS-CoV-2 Spike N501Y. Cat. No. 53-0780-96

- Eurofins developed a kit, the ViroBOAR SPIKE 1.0 RT-PCR Kit, used for simultaneous qualitative detection of SARS-CoV-2 S gene variants in codon 501 and 570 for discrimination of SARS-CoV-2 wildtype virus and strains UK and SA (genomic RNA) and already pretested positive by RT-PCR method.

! Caution, these solutions are an interesting temporary tool to screen positive samples (e.g. for selection for sequencing). However, the presence of one or two mutations is not enough to identify formally a variant, only sequencing can answer this question. !

New variants will arise over time and these solutions might become obsolete.

➢ Emergency Sequencing of SARS-CoV-2

If you need **urgent sequencing of positive samples**, please let us know (guillain.mikaty@pasteur.fr). Our laboratory has the capacity for a limited number of sequence and identification of variants.

MediLabSecure can cover all the fees involved!

➢ Sequencing sanger of SARS-CoV-2

For those of you equipped with **Sanger sequencers**, we produced a Standard Operating Procedure (SOP) that will help you to **sequence part or the whole of the genome of SARS-CoV-2.**

! The version attached is not yet validated experimentally in our laboratory !

We are in the process of validating all the primers of the S gene on SARS-CoV-2 strains in our laboratory. We will send you a validated SOP as soon as available!

You will find in this SOP a **complete list of primers used for RT-PCR and sequencing** that cover the whole genome of SARS-CoV-2. You can adapt your sequencing to any loci you want to target. We checked *in silico* that these primers should work on the current variants. However, there is a risk that future variant arise on the site of some of the primers… **!**

Let us know if we can help you implement the SOP in your laboratory.

➢ Sequencing MinION and training for SARS-CoV-2

For those of you equipped with **Oxford nanopore NGS sequencers MinION**, we will soon produce a Standard Operating Procedure (SOP) based on the Arctic (v3) procedure and share it with you to help you to sequence the whole of the genome of SARS-CoV-2.

If you need training for the sequencing of SARS-CoV-2 viruses with this technology, please contact us (guillain.mikaty@pasteur.fr).

-> We are currently working with the NIH that created an online training for this technology and we can try to help you access to the next session. Our colleagues from Tunis and Algeria already followed this training.

-> If the need is wide in the network, we might create a workshop hands-on on the MinION sequencing next spring in Paris.

➢ MediLabSecure Hotline on WhatsApp

I remind you that we created a **Hotline for diagnostic-related questions** within the network. This hotline use the WhatsApp platform, and has been working since April 2020. It has proved quite useful to quickly answer questions during the current crisis, even in the recent apparition of the variants.

We limit the hotline to one person per laboratory (two maximum) in order to keep the discussion simple and efficient. If your laboratory is not represented in the current group, do not hesitate to **ask us** to add you to it.

- **As always in MediLabSecure:**

❖ Support for diagnostics whenever needed. If you ever find yourself in need of help in a diagnostic, do not hesitate to send us a message. One of our mission as a network is to support each other! We can give a hand with technics you do not master or to attempt to identify an unknown disease.

❖ Support for reagents and protocols of relevant pathogens. In case of emergency, we are here to help!

❖ Experts'exchange: we can support the cost for an exchange of experts between two laboratories of the network (including with other specialties).

❖ Support for scientific publications (for partners publishing a scientific paper related to the MediLabSecure project).

❖ All Newsletters from MediLabSecure (General, entomology, human virology): https://www.medilabsecure.com/resources_newsletters.html

Kindest regards,

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