Respiratory Viruses in Luxembourg (ReViLux)

Weekly report (23 – 29 August 2021).

# Executive Summary

The Sentinel Surveillance Network identified one case of influenza-like illness, thus remaining below the recommended threshold for the interepidemic season, according to the European Center for Disease Prevention and Control (ECDC) guidelines.

Regarding SARS-CoV-2 genomic surveillance, the Laboratoire national de santé analysed 270 Luxembourgish specimens from week 34/2021 (from 542 total cases in the Grand Duchy of Luxembourg, 49,8%). This exceeds the minimum coverage (10%), but does not reach the minimum sample size (285) allowing to detect a 2.5% incidence, as recommended by the ECDC.

All specimens from week 34/2021 were assigned to Delta variant. Community surveillance showed that the parental lineage B.1.617.2 continues to be the most frequent one (85,7%), followed by AY.4 (10,5%). In respect to target group surveillance, all cases analysed, including 32 reinfection and post-vaccination breakthrough cases, were identified as Delta variant.

**Dominant variant**

Delta

**Sequencing Coverage**

49,8%

(

**SARS-CoV-2 positive cases**

542

# Introduction

The Laboratoire national de santé, as **National Reference Laboratory for Acute Respiratory Infections in Luxembourg**, performs close surveillance on respiratory viruses, with a special focus on SARS-CoV-2. There are currently three active projects:

**The Sentinel Surveillance Network**. It provides a broad picture of respiratory diseases affecting the Luxembourgish population, based on its double monitoring system (syndromic and virological).

**The National SARS-COV-2 Genomic Surveillance Program**. It enables detailed observation of SARS-CoV-2 mutations and variants through time and space, and also monitoring specific groups of interest.

**The COVVAC Serology Project**. It assesses the post-vaccination serological status in long-term care facilities and its evolution over time.

The ReViLux provides updates on the first two projects.

# Sentinel Surveillance Network

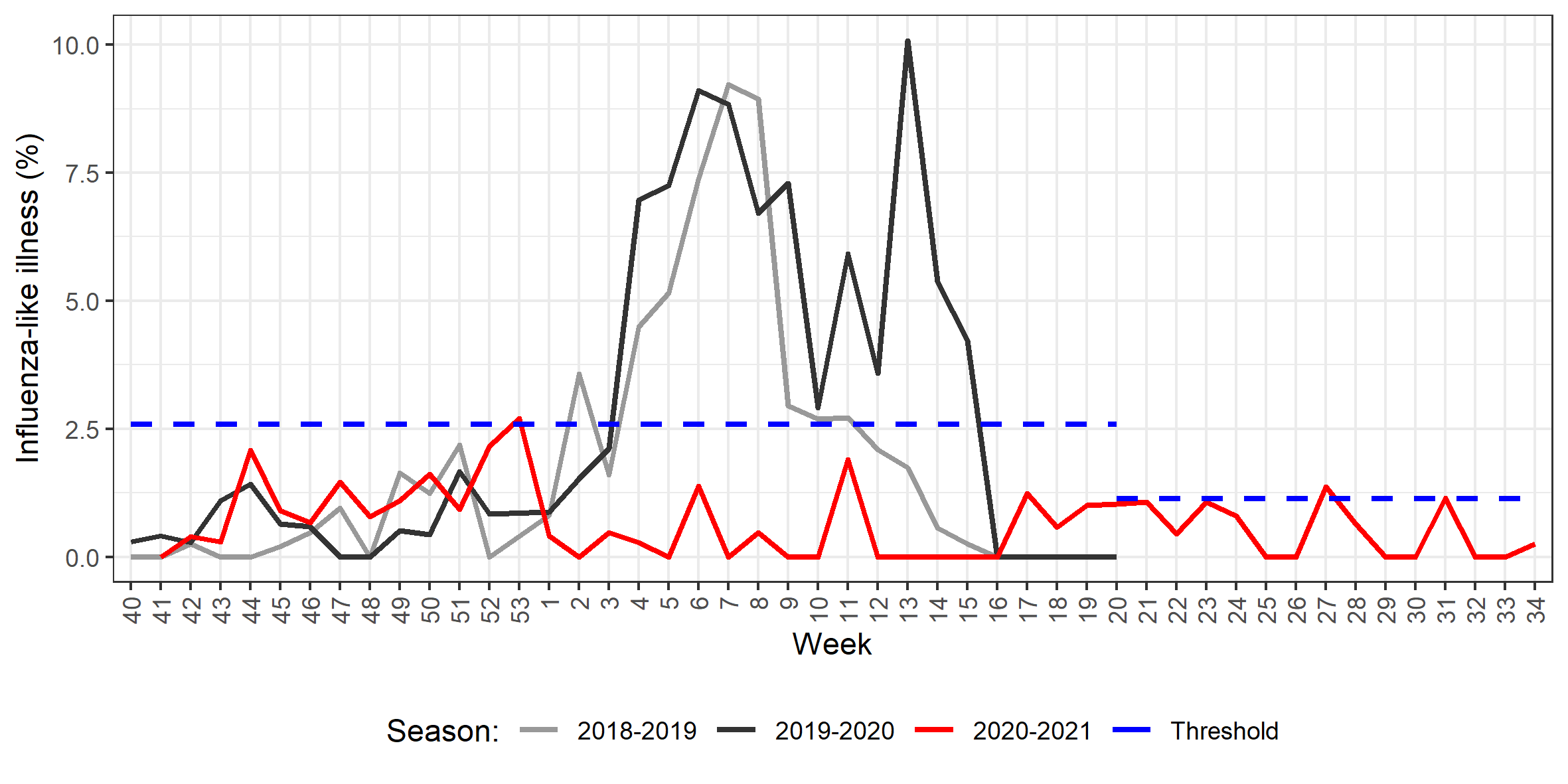
The **Sentinel Surveillance Network** aims at monitoring the circulating respiratory viruses, including SARS-CoV-2, and hence underpin public health actions. Following the World Health Organization (WHO) and European Centre for Disease Prevention and Control (ECDC) guidance, it focuses on cases of acute respiratory infection (ARI) and influenza-like illness (ILI).

Results of syndromic surveillance during week 34 are displayed in Table 1 and the history of ILI consultations since the 2018-2019 season is shown in Figure 1. One case of ILI was identified in week 34 (out of 394 consultations); therefore, **the percentage of ILI (0,25%) remains below the threshold for the interepidemic season** (1,14%), according to the ECDC.

Regarding the virological surveillance, no data is available for week 34.

*Table 1. Syndromic surveillance during week 34*

|  |  |  |  |  |  |
| --- | --- | --- | --- | --- | --- |
| Week | ARI | | ILI | | Total consultations |
| N | % | N | % |
| 2021/31 | 3 | 3.45% | 1 | 1.15% | 87 |
| 2021/32 | 15 | 8.47% | 0 | 0.00% | 177 |
| 2021/33 | 15 | 6.67% | 0 | 0.00% | 225 |
| 2021/34 | 20 | 5.08% | 1 | 0.25% | 394 |
| ARI: Acute Respiratory Infections (acute respiratory syndrome like bronchitis, pharyngitis, rhinitis, pneumonia… with or without fever). ILI: Influenza-Like Illness (acute respiratory syndrome <10 days, fever 38 ºC, systemic symptoms like myalgia or malaise…). | | | | | |



*Figure 1. Percentage of patients with influenza-like illness over the last three seasons*

# SARS-CoV-2 Genomic Surveillance

## The current sequencing strategy

The National Reference Laboratory for Acute Respiratory Infections at LNS receives SARS-CoV-2 -positive samples for (nasopharyngeal or oropharyngeal swabs analysed by RT-PCR) from the national network of laboratories and proceeds as follows:

1. Sequencing all specimens from hospital cases.
2. Sequencing all specimens from reinfection and post-vaccination cases.
3. Sequencing all specimens from cluster cases.
4. Sequencing a representative sample of community cases.

The representative sample of community cases is a systematic selection from all SARS-CoV-2 positive cases registered in Luxembourg to detect emerging variants and early increases in their incidence and transmission within the community in Luxembourg. This sample is selected according to the ECDC guidelines.

The LNS shares its sequencing results with GISAID EpiCov database (www.gisaid.org) periodically. SARS-CoV-2 lineages (variants) have been assigned based on Rambaut et al. using the Phylogenetic Assignment of Named Global Outbreak LINeages (pangolin) software (v3.1.11, pangoLEARN 2021-08-09). The ReViLux continues to use the Pango nomenclature, in addition to the WHO nomenclature, to allow easier visualization of links between any evolving variants and their ancestor (<https://cov-lineages.org>). See nomenclature equivalences in Annex 1.

**Methodological note**

Since 3 September 2021, the ECDC no longer considers B.1.1.7 and B.1.1.7+E484K lineages (Alpha variant) as variants of concern. This decision is based in both their low circulation and the high effectiveness of vaccines in controlling them.

Since 24 August 2021, the Phylogenetic Assignment of Named Global Outbreak Lineages (PANGOLIN) software uses a new version of its reference library (pango-designation v1.2.66), which includes 13 additional Delta lineages (AY.13 to AY.25). Both B.1.617.2 and AY lineages are classified as Delta variant by the World Health Organization.

## Sequenced specimens

Figure 2. Flowchart of specimens sequenced for week 34/2021

In week 34, 542 new cases were registered in Luxembourg; hence, the minimum sample size required to detect emerging variants at a 2.5% incidence is estimated to be 285 specimens (52,6%).

The microbial genomics unit at the LNS sequenced 285 specimens, with 270 specimens having been collected in week 34 from residents (49,8% coverage of the 542 total cases registered in Luxembourg; see coverage trend in Figure 2). This exceeds the minimum coverage (10%), but does not reach the minimum sample size (285) to detect a 2.5% incidence, as recommended by ECDC. The representative sample of community cases is based on a systematic selection.

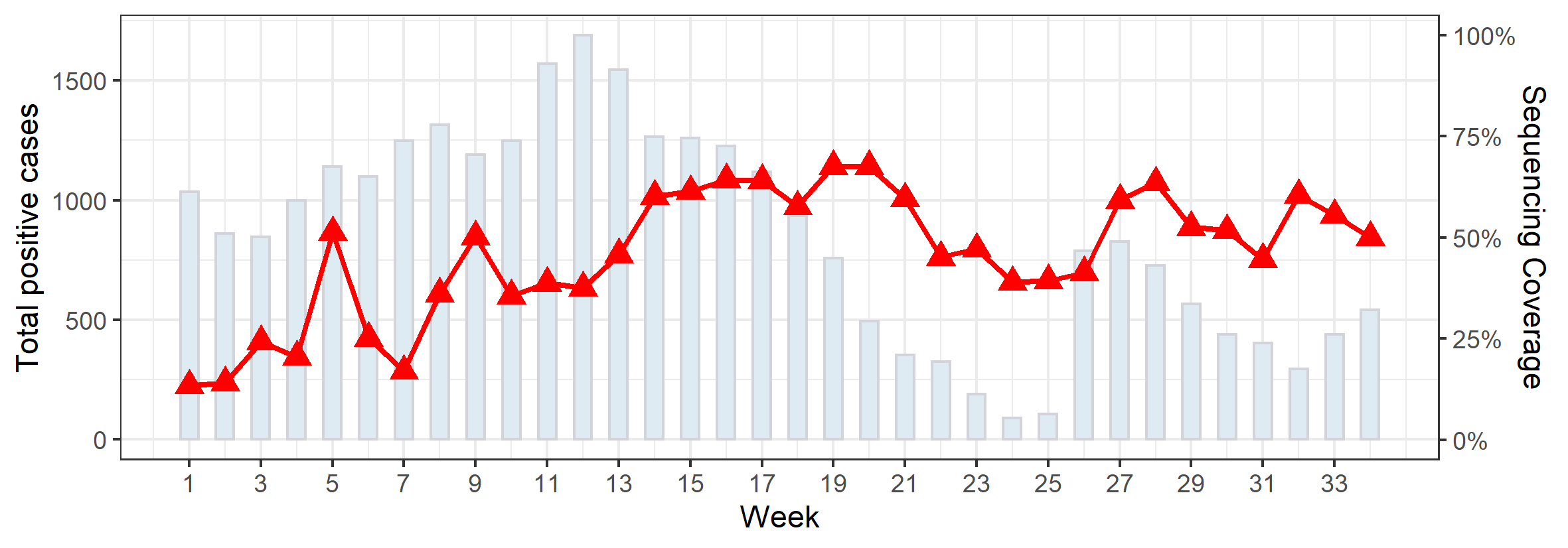


Figure 2. Sequence coverage based on weekly number of positive cases in Luxembourg during 2021

## Circulating lineage detection

The evolution of variants over the weeks is shown in Figure 3.

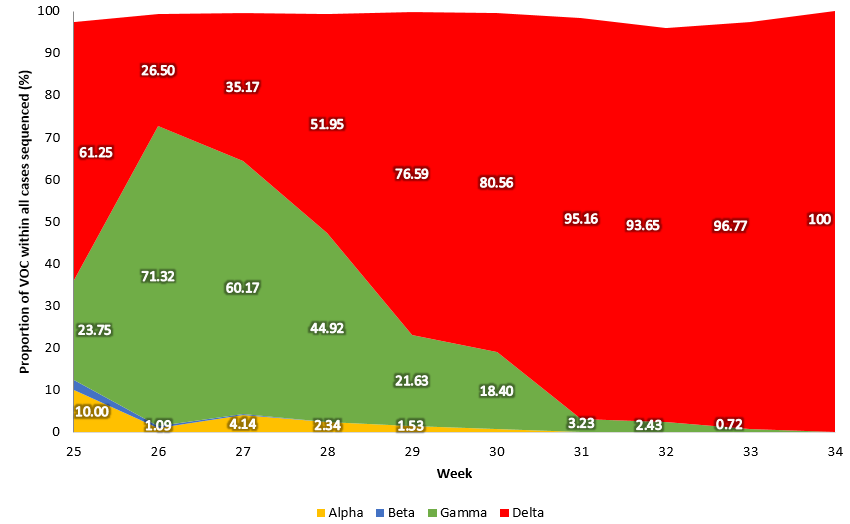


Figure 3. Evolution of VOCs in sequencing pool of all specimens including targeted sequencing (clusters, non-residents) over the last 10 weeks in Luxembourg

In week 34/2021, only Delta variant cases were detected within our population sequencing pool (after removal of cluster and non-resident specimens), including 5 Delta subtypes. The distribution of VOCs is displayed in Table 2, and their evolution over the weeks is shown in Figure 4.

*Table 2. Distribution of SARS-CoV-2 lineages detected within the community (cluster and non-resident cases excluded) in weeks 33 and 34/2021 (previous cases updated by retrospective sequencing)*

|  |  |  |  |  |  |  |
| --- | --- | --- | --- | --- | --- | --- |
| VOC | Week 33 | | | Week 34 | | |
| ***N*** | ***%*** | ***CI %*** | ***N*** | ***%*** | ***CI %*** |
| Delta B.1.617.2 | 186 | 84.5 | 79.8 - 89.3 | 226 | 85.3 | 81.0 - 89.5 |
| Delta AY.4 | 24 | 10.9 | 6.8 – 15.0 | 30 | 11.3 | 7.5 - 15.1 |
| Delta AY.12 | 6 | 2.7 | 0.6 - 4.9 | 4 | 1.5 | 0.0 – 3.0 |
| Delta AY.5 | 0 | 0.0 | 0.0 – 0.0 | 3 | 1.1 | 0.0 - 2.4 |
| Delta AY.6 | 2 | 0.9 | 0.0 - 2.2 | 2 | 0.8 | 0.0 - 1.8 |
| Gamma | 1 | 0.5 | 0.0 - 1.3 | 0 | 0.0 | - |
| Others | 0 | 0.0 | - | 0 | 0.0 | - |
| **Total** | **220** | **100.0** |  | **265** | **100.0** |  |

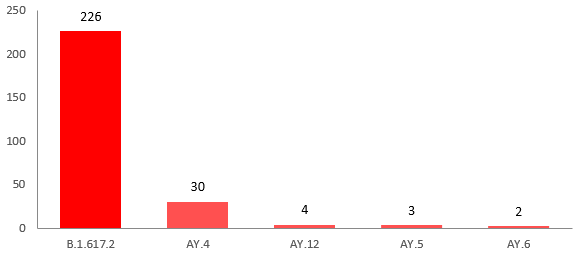


Figure 4. Number of SARS-CoV-2 variants in representative sample for week 34/2021

## Mutation surveillance

In addition to the surveillance of SARS-CoV-2 variants, the LNS monitors the occurrence of SARS-CoV-2 mutations assumed to have a clinical and epidemiological relevance.

Table 3 provides the overall frequencies of these mutations, detected in the lineage-assignable genome sequences, analyzed since 1 Sep 2020 (N = 17469), as well as the frequencies in week 34/2021.

*Table 3. Analysis of clinically relevant mutations identified during week 34/2021 sequencing*

|  |  |  |  |  |  |  |
| --- | --- | --- | --- | --- | --- | --- |
| Mutation | Gene | Genomic Position in reference | Frequency Overall [%] | Frequency Week 34/2021 [%] | Characteristics | Reference |
| D614G | S gene | 23402 | 95.8 | 100.0 | Higher infectivity, higher case fatality rate, higher transmission | Eaaswarkhanth 2020 Becerra-Flores 2020,  Hu 2020, Plante 2020 |
| P323L | ORF1ab | 14407 | 89.0 | 95.7 | Higher severity | Biswas & Mudi 2020 |
| R203K | N gene | 28880 | 53.5 | 0.0 | Fitness advantage for the virus | Leary 2020 |
| G204R | N gene | 28883 | 53.2 | 0.0 | Fitness advantage for the virus | Leary 2020 |
| N501Y | S gene | 23063 | 52.3 | 0.0 | 501Y.V1/V2; Improved ACE2 binding affinity/higher transmissibility | Filip Fratev 2020  COVID-19 Genomics Consortium UK, 2020 |
| E484K | S gene | 23012 | 15.5 | 0.0 | 501Y.V2 / possible impact on antibody neutralization activity (escape mutation), improved ACE2 binding affinity | Greaney 2020 |
| Y144del | S gene | 21991-21993 | 41.3 | 0.3 | possible impact on antibody binding affinity | Dawood 2020 |
| H69/V70del | S gene | 21765-21770 | 40.8 | 0.0 | possible impact on antibody neutralization activity and reinfection; included in "mink" mutation | Kemp 2020 |
| P681H | S gene | 23604 | 38.7 | 0.3 | immediately adjacent to the furin cleavage site, a known location of biological significance | COVID-19 Genomics Consortium UK, 2020 |
| L37F | Nsp6 | 11081 | 3.4 | 5.0 | Favored viral infection, higher severity | Aiewsakun 2020 |
| Q57H | ORF3a | 25561 | 19.6 | 0.0 | Higher severity | Biswas & Mudi 2020 |
| K417N | S gene | 22813 | 7.0 | 0.0 | 501Y.V2 / possible impact on antibody binding affinity (escape mutation) | Kemp 2020 |
| N439K | S gene | 26143 | 0.7 | 0.0 | Improved ACE2 binding affinity | Zhou 2020 |

# References

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Rambaut A., Holmes E., O’Toole Á., Hill V., McCrone J., Ruis C. et al. (2020). A dynamic nomenclature proposal for SARS-CoV-2 lineages to assist genomic epidemiology. Nature Microbiology, 5(11), 1403-1407. doi: 10.1038/s41564-020-0770-5

# Annexes

## Annex 1. SARS-CoV-2 variants naming

The ReViLux continues to use the Pango nomenclature, in addition to the WHO nomenclature, to allow easier visualization of links between any evolving variants and their ancestor. Equivalence for VOC are shown in Table A1 (adapted from WHO).

*Table A1. Nomenclature for variants of concern by the World Health Organization (WHO)*

|  |  |  |  |  |  |  |
| --- | --- | --- | --- | --- | --- | --- |
| WHO label | Pango lineage\* | GISAID clade/lineage | Nextstrain clade | Additional amino acid changes monitored | Earliest documented samples | Date of designation |
| Alpha | B.1.1.7# | GRY (formerly GR/501Y.V1) | 20I (V1) | +S:484K  +S:452R | United Kingdom, Sep-2020 | 18-Dec-2020 |
| Beta | B.1.351 | GH/501Y.V2 | 20H (V2) | +S:L18F | South Africa, May-2020 | 18-Dec-2020 |
| Gamma | P.1 | GR/501Y.V3 | 20J (V3) | +S:681H | Brazil, Nov-2020 | 11-Jan-2021 |
| Delta | B.1.617.2§ | G/478K.V1 | 21A | +S:417N | India, Oct-2020 | VOI: 4-Apr-2021  VOC: 11-May-2021 |

*\*All sublineages included. # includes all Q sublineages.* § *includes all AY sublineages.*

*Adapted from World Health Organization - Tracking SARS-CoV-2 variants*