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EPIDEMIOLOGY OF INFECTIOUS DISEASES
TRANSVERSAL ACTIVITIES IN APPLIED GENOMICS
FOODBORNE PATHOGENS

WASTEWATER-BASED EPIDEMIOLOGICAL SURVEILLANCE

Weekly report
2025 week 09

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Sciensano can count on more than 950 staff members who are committed to health every day.
As our name suggests, science and health are central to our mission. Sciensano's strength and uniqueness lie within the holistic and multidisciplinary approach to health. More particularly we focus on the close and indissoluble interconnection between human and animal health and their environment (the "One health" concept). By combining different research perspectives within this framework, Sciensano contributes in a unique way to everybody's health.
For this, Sciensano builds on the more than 100 years of scientific expertise.

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National wastewater-based epidemiological surveillance

February 2025 • Brussels • Belgium
Reference number: D/2024.14.440/28 • ISSN 2795-7152

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With the financial support of

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Please cite as: R. Janssens, H. Maloux, A. Meyers, M. Lesenfants, L. Van Poelvoorde, N. Roosens, B. Verhaegen, J. Linussio, K. Van Hoorde, S. Van Gucht, K. De Ridder, K. Blot, V. Hutse. Wastewater-based epidemiological surveillance – Weekly report. Brussels, Belgium : Sciensano ; 2025 26p. Report number: D/2024.14.440/28 ISSN 2795-7152. Available from: https://www.sciensano.be/en/biblio/wastewater-based-epidemiological-surveillance-weekly-report

1. TABLE OF CONTENTS

2. KEY POINTS

The epidemiological situation of several respiratory viruses measured through wastewater samples is presented in this document. Samples are collected once a week in 30 wastewater treatment plants (WWTPs) covering a total of 38% of the Belgian population. Additionally samples from the treatment plant of Brussels Airport are also collected for the genomic surveillance.

The definition of an area in alert depends on the respiratory virus. For SARS-CoV-2, an area is in alert if the threshold for the High Circulation indicator is reached. For RSV and influenza, an area is in alert if the virus is detected.

Conclusions based on the latest results dating of week 09 (24-02-2025):

- **Overview of respiratory viruses:** RSV and influenza are detected in Belgian wastewaters. The number of areas with at least one respiratory virus in alert is 25, over 30 areas.
- **SARS-CoV-2**
  - **National level**: SARS-CoV-2 is at a low level compared to the wave starting in November 2023. The number of areas above the threshold for the High Circulation and Increasing trend indicators is respectively 1 and 1, over 30 areas.
  - **Regional level**: The situation is similar to the national one in all regions.
  - **Genomic surveillance**: During the wave starting in September 2024, the KP.3 variant was dominant.
- **RSV**: The number of areas where RSV is detected is at a high level. The number of areas where RSVA or RSVB is detected is 16, over 30 areas.
- **Influenza**: The number of areas where influenza is detected, is at a very high level. The number of areas where influenza A or B is detected is 23, over 30 areas.

The wastewater situation can be followed through:

- The weekly bulletin on respiratory infections published in French and Dutch
- The wastewater respiratory viruses dashboard
- Methodology available in the Methodology Appendix
- General information about the surveillance available on the website of Sciensano
- Data are available on the Belgian federal geoportal

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3. INTRODUCTION

The national wastewater-based epidemiological surveillance of COVID-19 started in September 2020. In January 2024, respiratory syncytial virus (RSV) and influenza (Flu) were integrated in this surveillance. The present report aims to assess the wastewater-based epidemiological situation in Belgium for those respiratory viruses. This report is updated weekly on Monday with results based on samples collected the previous Monday.

Concentrations are measured once a week in 30 wastewater treatment plants (WWTPs). The epidemiological situation of SARS-CoV-2 is assessed on the basis of two wastewater-based indicators, defined in the methodology section (see section 4.3.1). The assessment is performed at different spatial levels: national, regional, provincial and for the areas covered by the treatment plants. Regarding RSV and influenza, the situation is assessed through the number of areas in which viruses were detected.

The genomic SARS-CoV-2 surveillance is performed at surges and peaks of viral circulation, based on samples from four treatment plants: Brussels-North, Gent, Liège-Oupeye, and Brussels Airport. Sequencing of wastewater samples provides information on the viral genomic diversity circulating in the general population. Genomic results are presented in this report only if recent analyses were performed. Otherwise, the date of the last publication with genomic results is mentioned.

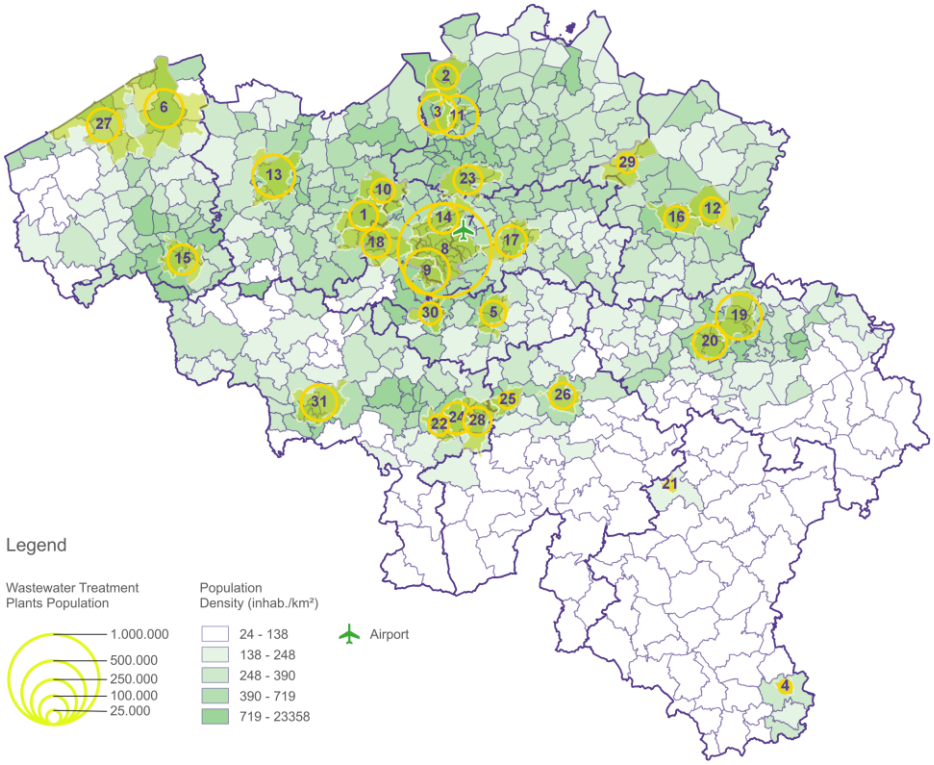
4. METHODOLOGY

4.1. Sample collection and analysis

Samples are collected once a week in several wastewater treatment plants (WWTP) covering around 38% of the Belgian population: 30% in the Flemish region, 30% in the Walloon region, and nearly 100% in the Brussels region. Figure 1 shows the catchment areas covered by the WWTP located in areas with high population density. The catchment area of a WWTP corresponds to the geographical area from which the wastewater are collected. Additionally samples from the treatment plant of Brussels Airport are also collected for the genomic surveillance.

Samples are collected on Mondays by auto-samplers (24-hour composite) at the influent of WWTP. They are then transported to two laboratories for quantification of SARS-CoV-2, RSV, influenza and Pepper Mild Mottle Virus (PMMoV). Results are made publicly available on the following Monday.

Further details on the coverage, sampling plan, and analytical method can be found in the Methodology Appendix



WWTP	ID	Population
Aalst	1	98357
Antwerpen-Noord	2	73974
Antwerpen-Zuid	3	178677
Arlon	4	19440
Basse Wavre (Dyle)	5	78290
Brugge	6	180586
Brussels Airport	7	Na
Bruxelles-Nord	8	1080145
Bruxelles-Sud	9	249307
Dendermonde	10	69500
Deurne	11	215592
Genk	12	68582
Gent	13	220130
Grimbergen	14	107643
Harelbeke	15	107894
Hasselt	16	69586
Leuven	17	120939
Liedekerke	18	101075
Liège Oupeye	19	253780
Liège Sclessin	20	142240
Marche-en-Famenne	21	8633
Marchienne-au-Pont	22	52017
Mechelen-Noord	23	98143
Montignies-sur-Sambre	24	123576
Mornimont	25	34139
Namur-Brumagne	26	82175
Oostende	27	135491
Roselies	28	96890
Tessenderlo	29	45125
Vallée du Hain (l'Orchis)	30	57826
Wasmuel	31	157343

Figure 1 ● Population located in areas covered by the wastewater treatment plants (highlighted in yellow) and population density for each municipality (indicated by the green scale).

4.2. Wastewater results

4.2.1. SARS-CoV-2

The concentration of SARS-CoV-2 in a sample is affected by rain events and population mobility. Therefore, a human maker called PMMoV is used to account for these effects. PMMoV is a well-known virus excreted by humans. Concentrations of SARS-CoV-2 and PMMoV are thus measured in the same

sample and divided to obtain the viral to faecal ratio expressed in copies of SARS-CoV-2 over copies of PMMoV.

Limit of quantification (LOQ) and limit of detection (LOD) of SARS-COV-2 were respectively estimated at 10 and 2 copies/ml. As PMMoV concentration is independent of SARS-CoV-2 concentration, a mean LOQ was imputed for the viral to faecal ratio when SARS-CoV-2 concentrations are below LOQ. The mean viral to faecal ratio obtained at a SARS-CoV-2 concentration of 10 c/ml is 200 10e-6 copies/copies.

Aggregation at national, regional, and provincial level are computed using the mean viral to faecal ratio weighted by the population covered in each area.

4.2.2. RSV and Influenza

LOQ for RSV and influenza is estimated at 25 copies/ml. As RSV and influenza concentrations remains below the estimated LOQ during most of the year, the situation is assessed based on the detection or absence of detection rather than using quantitative results.

4.3. Alerting indicators

4.3.1. SARS-CoV-2

Viral to faecal ratios should not be directly compared between catchment areas, provinces, nor regions as treatment plants and sewage systems have different characteristics. Despite application of the same protocols, slight differences can occur between laboratories. To mitigate these biases, two indicators are computed on normalized viral to faecal ratios, allowing for comparison between different areas:

- The **High Circulation** indicator highlights the catchment areas where the viral to faecal ratio is high. It corresponds to a situation where the viral to faecal ratios exceeds 50% of the highest value recorded during the wave starting in November 2023 (i.e. from 20/11/2023 to 01/01/2024).
- The **Increasing Trend** indicator highlights the catchment areas where the viral to faecal ratio has been increasing for more than 13 days. The indicator is computed based on the viral to faecal ratio moving average on the past two weeks.

The indicators were developed to provide information of different natures. The **Increasing Trend** indicator assesses the evolution over time, while the **High Circulation** indicator informs on the level of the current concentration. Indicators are computed at several levels: national, regional, provincial and catchment area.

4.3.2. RSV and influenza

The RSV and influenza's epidemiological situations are assessed once a week at a national level based on the total number of areas in which the viruses are detected.

4.3.3. Missing value

A missing value for the analysis of a virus can be caused by several reasons: i) impossibility to sample, ii) transport issue or iii) impossibility to analyse a sample due to PCR inhibition or equipment failure. In case a missing value occurs for SARS-CoV-2 or PMMoV analysis, indicators are not computed and are set to missing. The list of areas with missing value can be found in Table A.1.

4.4. Genomic analysis of SARS-CoV-2

Genomic surveillance of SARS-CoV-2 is performed at surges and at peaks of circulation. Therefore, genomic results are presented in this report only if recent analyses were performed. Otherwise, the date of the last publication with genomic results is mentioned in the results section. Areas selected to conduct

sequencing are located in the three Belgian regions: Brussels-North, Gent, Liège-Oupeye, and Brussels Airport. Brussels Airport is an important entry point for variants on the Belgian territory.

Variant of concern (VOC) and variant of interest (VOI) as defined by the ECDC classification system are presented in the results section. Variant under monitoring (VUM) are grouped in a category named “Other”. And, variant not belonging to the ECDC classification system are gathered in a category named “Unassigned by ECDC”. Further details on the classification and analytical method are available online.

5. RESULTS

5.1. Overview of respiratory viruses

Figure 2 presents the evolution of the number of areas in alert for at least one respiratory virus. The definition of an area in alert depends on the virus. For SARS-CoV-2, an area is in alert if the threshold for the High Circulation indicator is reached. For RSV, an area is in alert if RSVA or RSVB is detected. For influenza, an area is in alert if influenza A or influenza B is detected. Table 1 represents the number of areas in alert for at least one respiratory virus in the last 10 weeks. Figure 3 presents the number of areas in alert for each virus.

RSV and influenza are detected in Belgian wastewaters. The number of areas with at least one respiratory virus in alert is 25, over 30 areas.

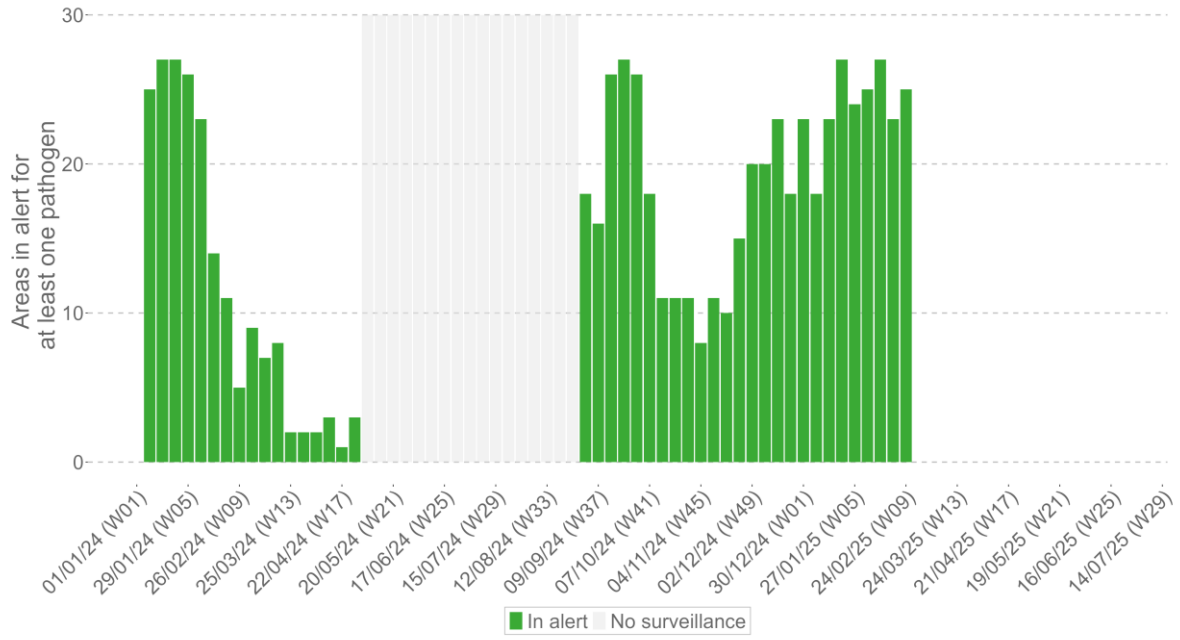


Figure 2 ● Total number of areas in alert (green bars) for at least one respiratory virus. The RSV and influenza surveillances are suspended during the summer period (grey bars).

Table 1 ● Total number of areas in alert for at least one respiratory virus in the last 10 weeks. Dates with a missing data are indicated with a "/".

Date	Areas in alert
2024-W52	18
2025-W01	23
2025-W02	18
2025-W03	23
2025-W04	27
2025-W05	24
2025-W06	25
2025-W07	27

Date	Areas in alert
2025-W08	23
2025-W09	25

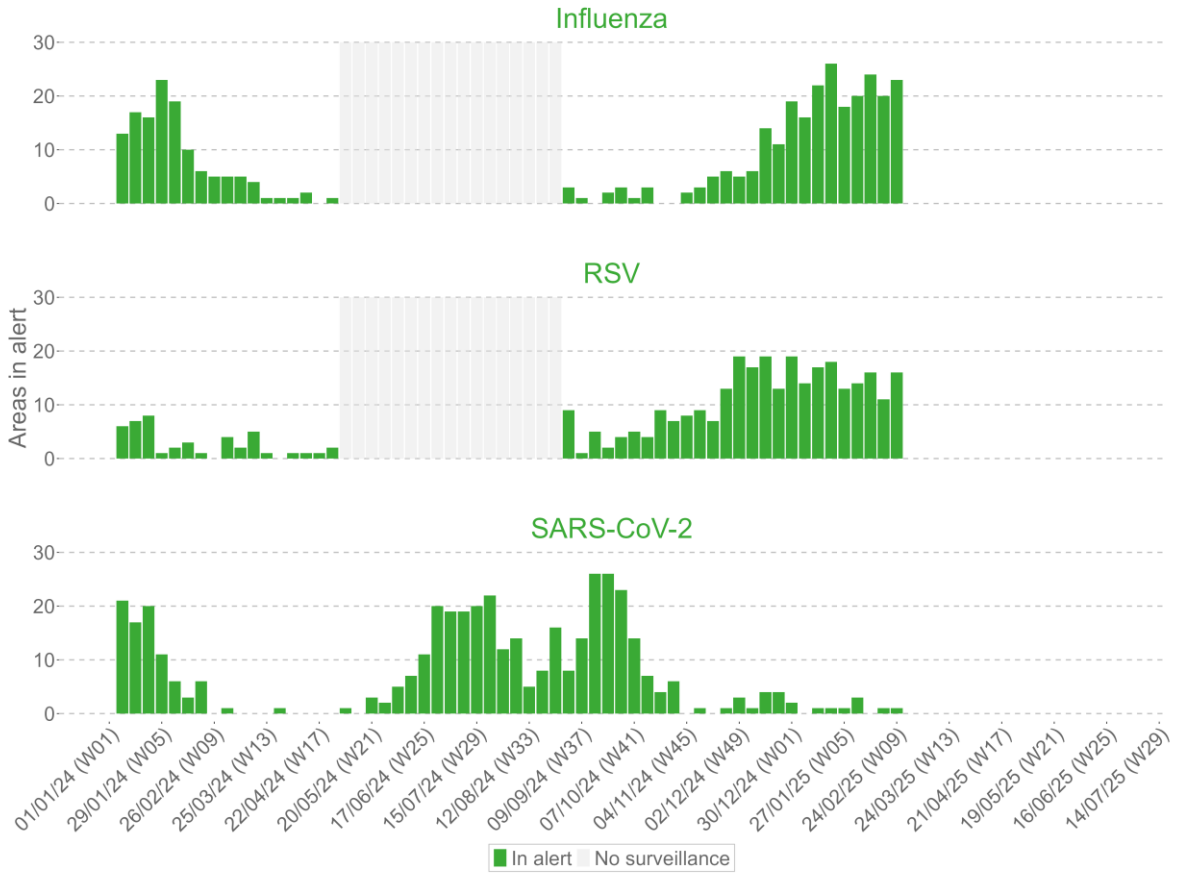


Figure 3 ● Total number of areas in alert by respiratory virus. Influenza and RSV surveillances are suspended during the summer period (grey bars).

5.2. SARS-CoV-2

5.2.1. National level

Figure 4 presents the SARS-CoV-2 viral to faecal concentration at a national level. Table 2 presents the national indicators for the last 10 weeks. Figure 5 presents on a map the indicators at area level. In Figure 6, the number of areas with indicator above threshold is presented over time.

SARS-CoV-2 is at a low level compared to the wave starting in November 2023. The number of areas above the threshold for the High Circulation and Increasing trend indicators is respectively 1 and 1, over 30 areas.

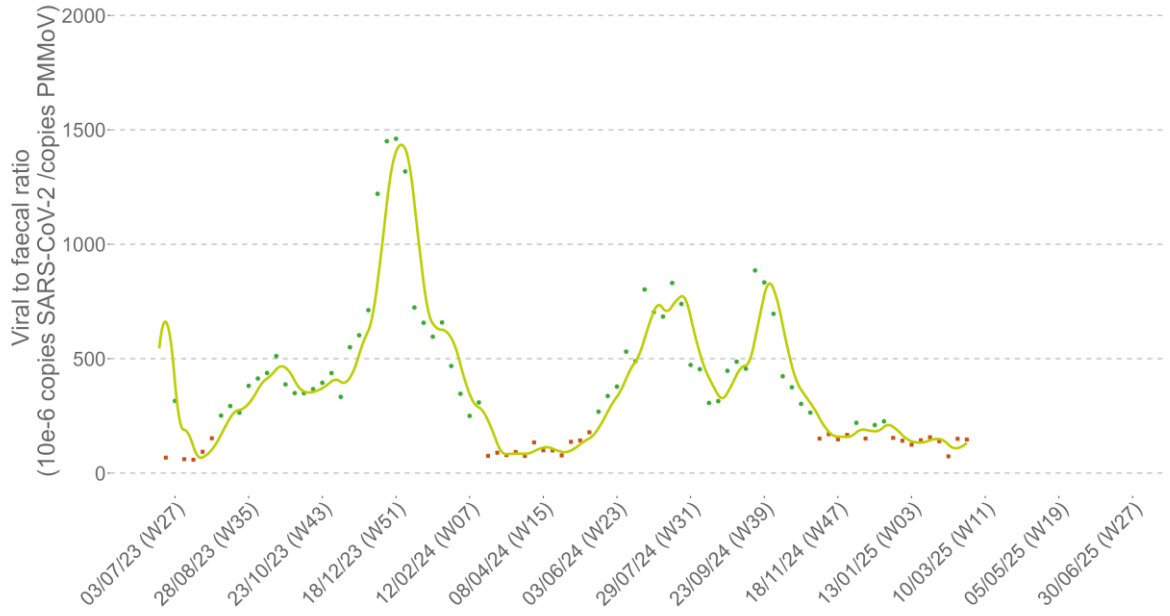


Figure 4 ● SARS-CoV-2 viral to faecal concentrations above LOQ (green dots) and below LOQ (red square) expressed in 10e-6 SARS-CoV-2 copies/ PMMoV copies and past two weeks moving average (yellow line).

Table 2 ● Indicators above threshold (1) or not (0) at the national level in the last 10 weeks. An indicator is displayed in bold if it was 0 last week and is 1 this week. Missing data is indicated with a "/" and data below LOQ is indicated with "bLOQ".

Date	High Circulation	Increasing Trend	Normalized viral ratio (%)	Mean viral ratio (10e-6)	Consecutive days of increase
2024-W52	0	0	15	226	7
2025-W01	0	0	bLOQ	bLOQ	0
2025-W02	0	0	bLOQ	bLOQ	0
2025-W03	0	0	bLOQ	bLOQ	0
2025-W04	0	0	bLOQ	bLOQ	0
2025-W05	0	0	bLOQ	bLOQ	0
2025-W06	0	0	bLOQ	bLOQ	0
2025-W07	0	0	bLOQ	bLOQ	0

Date	High Circulation	Increasing Trend	Normalized viral ratio (%)	Mean viral ratio (10e-6)	Consecutive days of increase
2025-W08	0	0	bLOQ	bLOQ	0
2025-W09	0	0	bLOQ	bLOQ	0

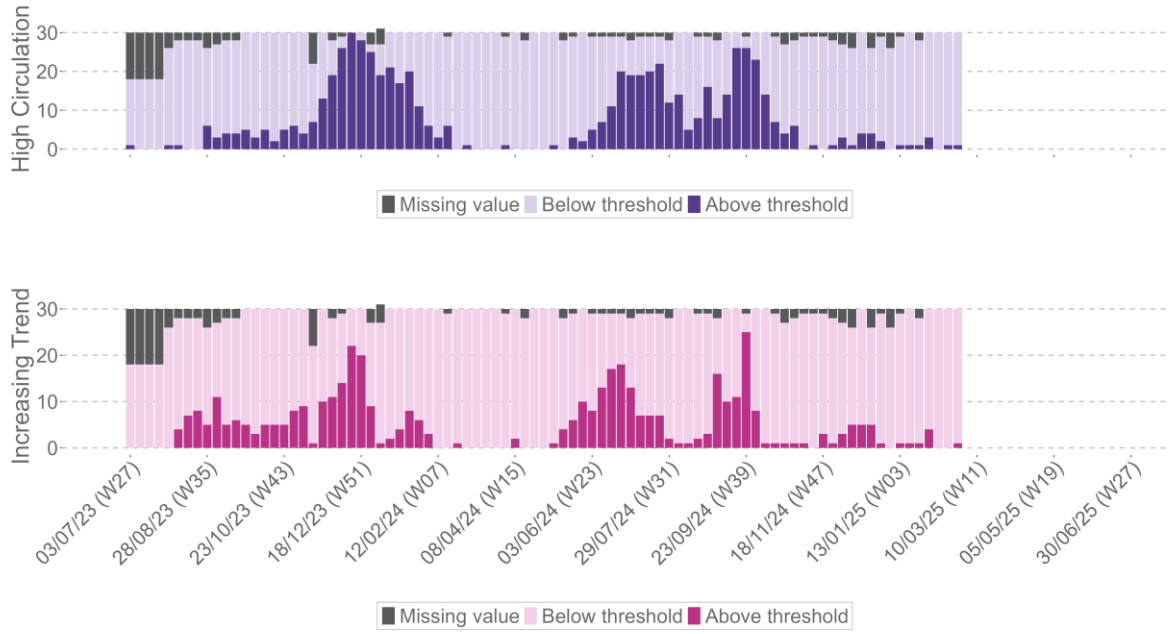


Figure 5 ● Number of areas having an indicator above threshold (coloured bars), below threshold (light coloured bars), and a missing indicator (grey bars).

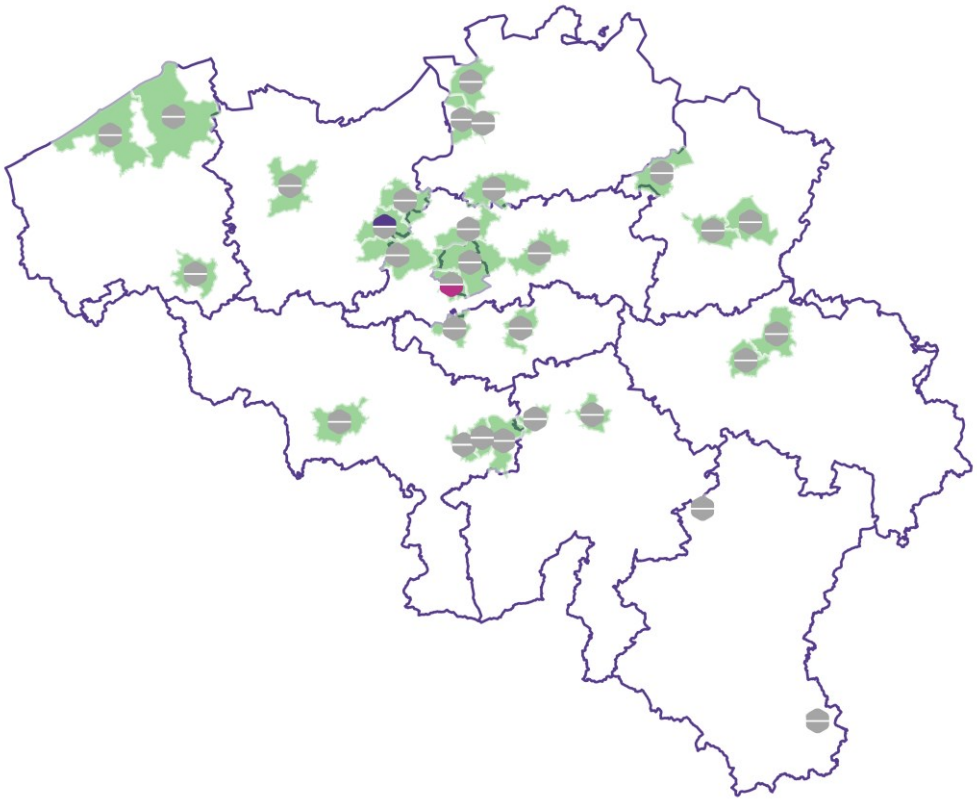


Figure 6 ● Geographical location of covered areas with corresponding status of the two indicators being above (colored pies) or below (grey pies) the threshold: High Circulation (in purple) and Increasing Trend (in pink). Missing data for an area are displayed in white color. Names of covered areas with respect of their localization can be found in Figure 1

5.2.2. Regional level

Figure 7 presents SARS-CoV-2 viral to faecal concentration at a regional level. Table 3 presents the indicators at a regional level.

The situation is similar to the national one in all regions.

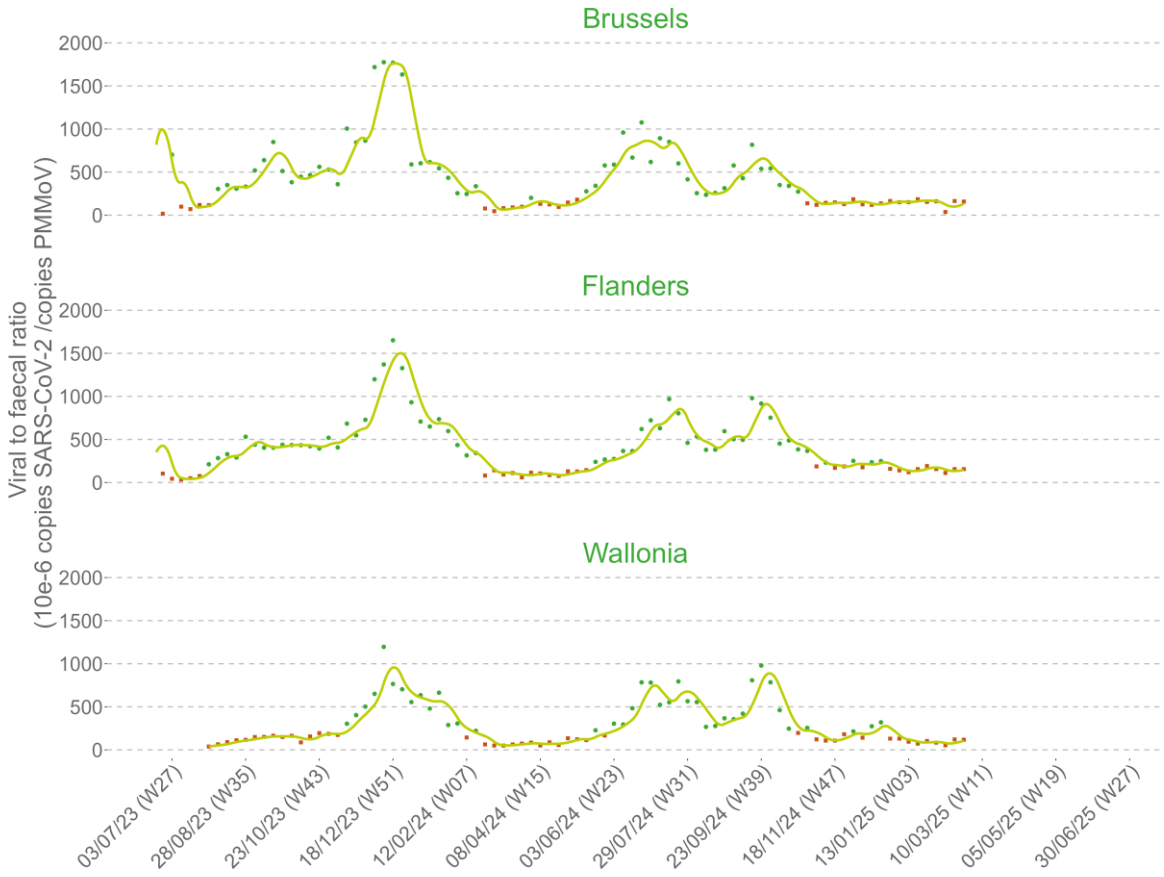


Figure 7 ● SARS-CoV-2 viral to faecal concentrations above LOQ (green dots) and below LOQ (red squares) expressed in 10e-6 SARS-CoV-2 copies / PMMoV copies and past two weeks moving average (yellow line).

Table 3 ● Indicators above threshold (1) or not (0) at the regional level. Indicators with a different value from last week, are displayed in bold. Missing data is indicated with a "/" and data below LOQ is indicated with "bLOQ".

Region	High Circulation	Increasing Trend	Normalized viral ratio (%)	Mean viral ratio (10e-6)	Consecutive days of increase
Brussels	0	0	bLOQ	bLOQ	0
Flanders	0	0	bLOQ	bLOQ	0
Wallonia	0	0	bLOQ	bLOQ	0

5.2.3. Provincial level

Table 4 presents the indicators at the provincial level.

Table 4 ● Indicators above threshold (1) or not (0) at the provincial level. Indicators with a different value from last week, are displayed in bold. Missing data is indicated with a "/" and data below LOQ is indicated with "bLOQ".

Province	High Circulation	Increasing Trend	Normalized viral ratio (%)	Mean viral ratio (10e-6)	Consecutive days of increase
Antwerpen	0	0	bLOQ	bLOQ	0
Brabant Wallon	0	0	bLOQ	bLOQ	0
Brussels	0	0	bLOQ	bLOQ	0
Hainaut	0	0	bLOQ	bLOQ	0
Liege	0	0	bLOQ	bLOQ	0
Limburg	0	0	bLOQ	bLOQ	0
Luxembourg	0	0	bLOQ	bLOQ	0
Namur	0	0	bLOQ	bLOQ	0
Oost-Vlaanderen	0	0	26	295	7
Vlaams-Brabant	0	0	bLOQ	bLOQ	0
West-Vlaanderen	0	0	bLOQ	bLOQ	0

5.2.4. Individual area level

Table 5 presents indicators at the individual area level.

Table 5 ● Indicators above threshold (1) or not (0) at the individual area level. Indicators with a different value from last week, are displayed in bold. Missing data is indicated with a "/" and data below LOQ is indicated with "bLOQ".

Area	High Circulation	Increasing Trend	Normalized viral ratio (%)	Mean viral ratio (10e-6)	Consecutive days of increase
Aalst	1	0	55	756	7
Antwerpen-Noord	0	0	bLOQ	bLOQ	0
Antwerpen-Zuid	0	0	bLOQ	bLOQ	0
Arlon	0	0	bLOQ	bLOQ	0
Basse Wavre (Dyle)	0	0	bLOQ	bLOQ	0
Brugge	0	0	bLOQ	bLOQ	0
Brussels-North	0	0	bLOQ	bLOQ	0
Brussels-South	0	**1**	8	208	14
Dendermonde	0	0	14	242	7
Deurne	0	0	bLOQ	bLOQ	0
Genk	0	0	bLOQ	bLOQ	0
Gent	0	0	bLOQ	bLOQ	0
Grimbergen	0	0	bLOQ	bLOQ	0
Harelbeke	0	0	bLOQ	bLOQ	0
Hasselt	0	0	7	290	7
Leuven	0	0	bLOQ	bLOQ	0
Liedekerke	0	0	bLOQ	bLOQ	0
Liege Oupeye	0	0	bLOQ	bLOQ	0
Liege Sclessin	0	0	17	220	7
Marche-en-Famenne	0	0	bLOQ	bLOQ	0
Marchienne-au-Pont	0	0	bLOQ	bLOQ	0
Mechelen-Noord	0	0	bLOQ	bLOQ	0
Montignies-sur-Sambre	0	0	bLOQ	bLOQ	0
Mornimont	0	0	bLOQ	bLOQ	0
Namur-Brumagne	0	0	bLOQ	bLOQ	0
Oostende	0	0	bLOQ	bLOQ	0
Roselies	0	0	bLOQ	bLOQ	0
Tessenderlo	0	0	bLOQ	bLOQ	0
Vallee du Hain (L'Orchis)	0	0	bLOQ	bLOQ	0
Wasmuel	0	0	bLOQ	bLOQ	0

5.3. RSV

The number of areas where RSV is detected is at a high level. The number of areas where RSVA or RSVB is detected is 16 over 30 areas, as presented in Figure 3. The detailed numbers for RSVA and RSVB are presented in Figure 8.

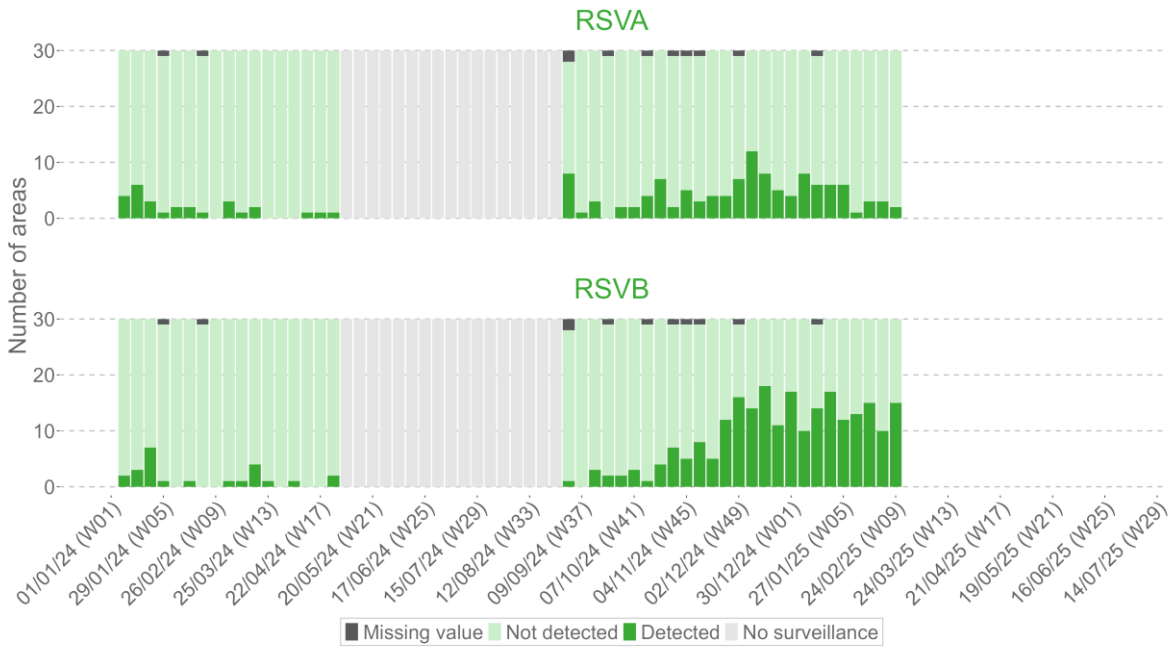


Figure 8 ● Number of areas with RSV detected (coloured bars), not detected (light coloured bars), and with a missing value (dark grey bars). RSV surveillance is suspended during the summer period (light grey bars).

5.4. Influenza

The number of areas where influenza is detected, is at a very high level. The number of areas where influenza A or B is detected is 23 over 30 areas, as presented in Figure 3. The detailed numbers for FluA and FluB are presented in Figure 9.

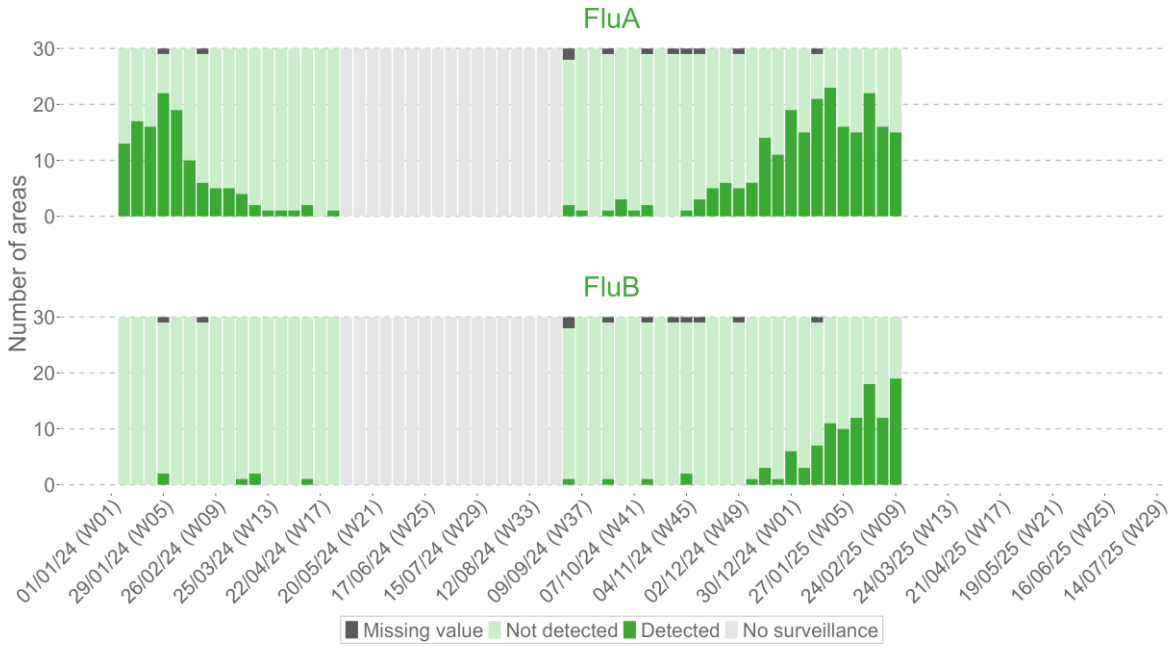


Figure 9 ● Number of areas with influenza detected (coloured bars), not detected (light coloured bars) and with a missing value (light grey bars). Influenza surveillance is suspended during the summer period (light grey bars).

5.5. Genomic surveillance of SARS-CoV-2

During the wave starting in September 2024, the KP.3 variant was dominant.
The viral ratios and variant proportions for the areas of Brussels-North, Gent, Liège Oupeye and Brussels Airport are shown in Figures 10 to 13, respectively. Information on the Pango lineages are presented in Table A.2.

Assessment of the situation can be found in the Summary section above.

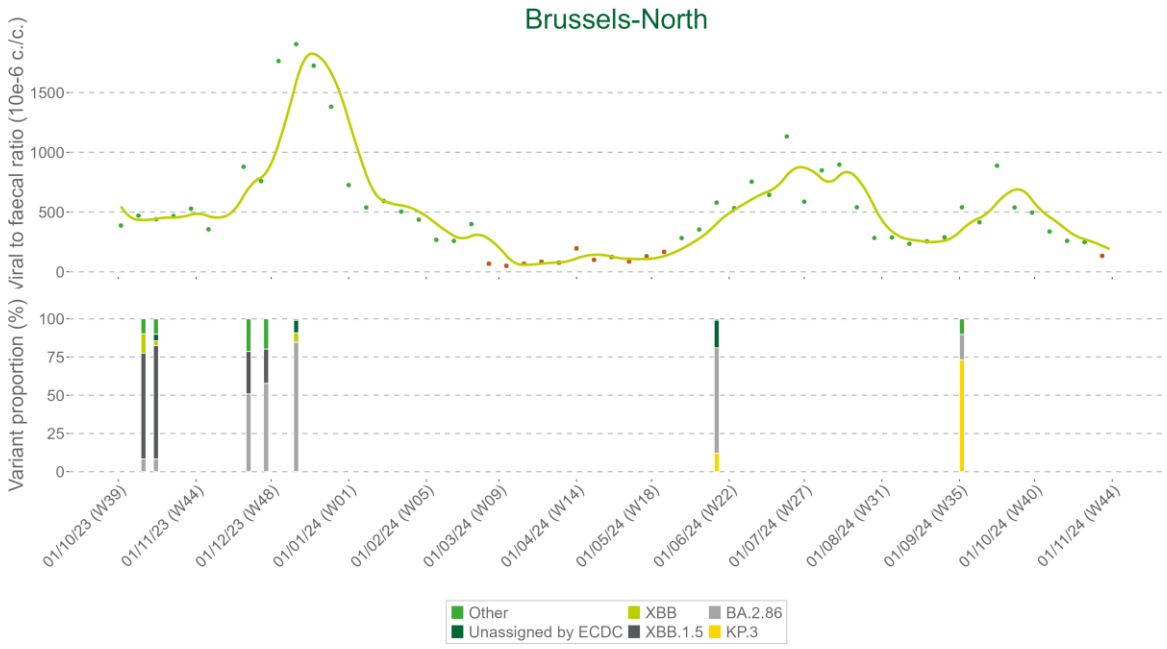


Figure 10 ● SARS-CoV-2 viral ratios expressed as SARS-CoV-2 copies / PMMoV copies (based on the past two weeks moving average) and variant proportions using the ECDC classification for the area of Brussels-North.

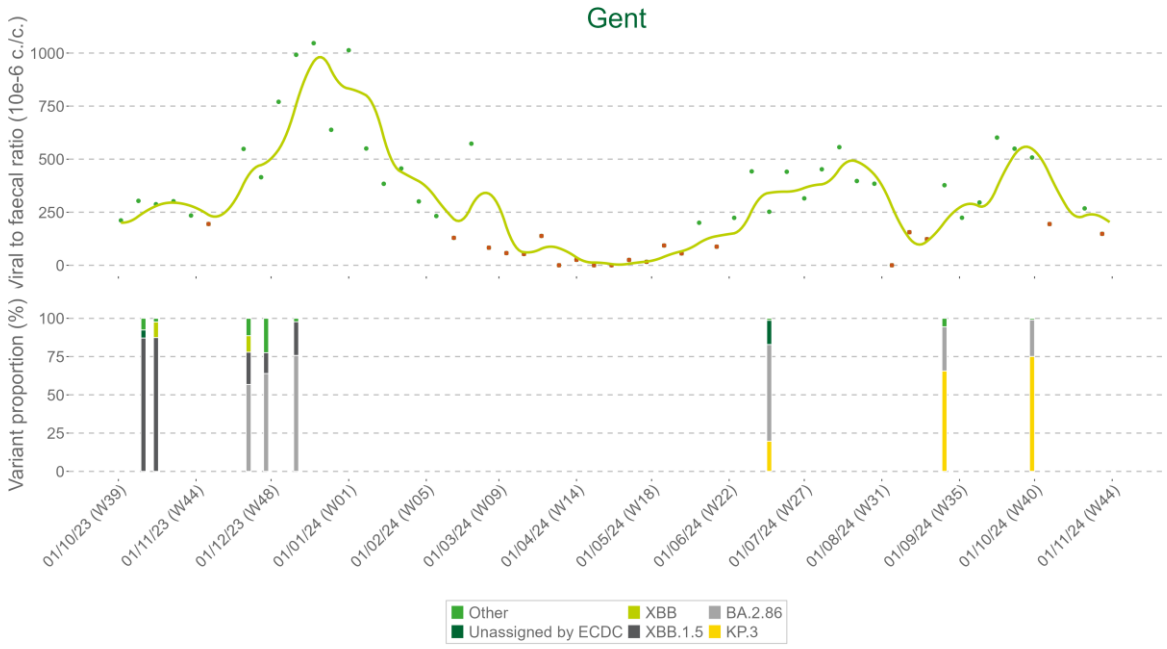


Figure 11 ● SARS-CoV-2 viral ratios expressed as SARS-CoV-2 copies / PMMoV copies (based on the past two weeks moving average) and variant proportions using the ECDC classification for the area of Gent.

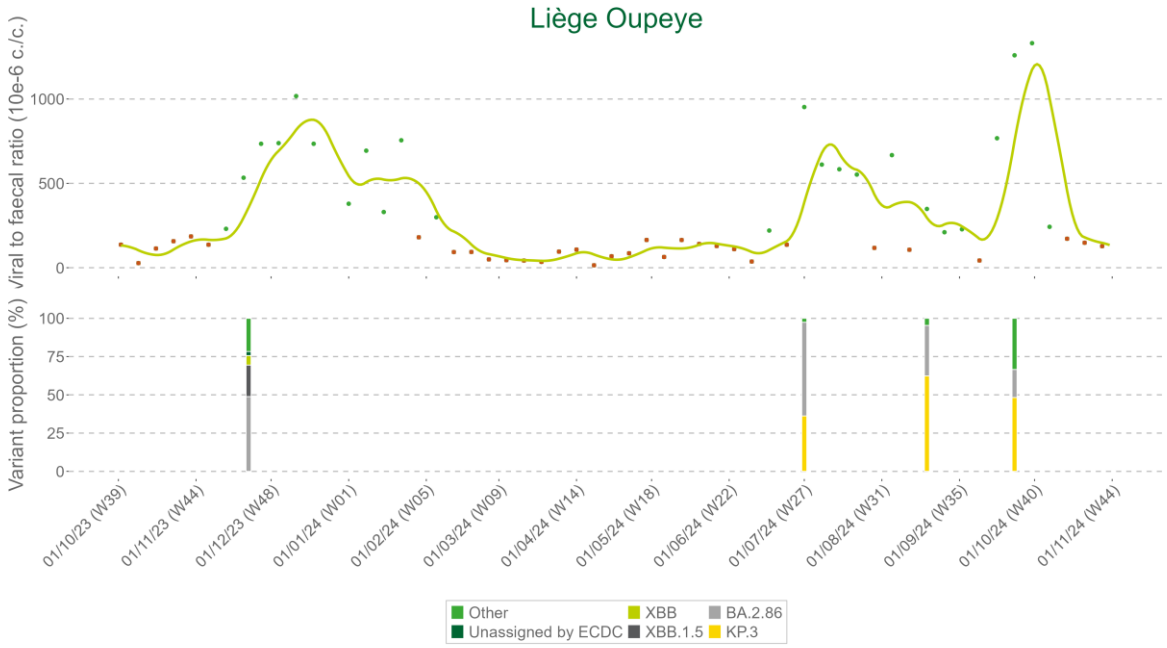


Figure 12 ● SARS-CoV-2 viral ratios expressed as SARS-CoV-2 copies / PMMoV copies (based on the past two weeks moving average) and variant proportions using the ECDC classification for the area of Liège Oupeye.

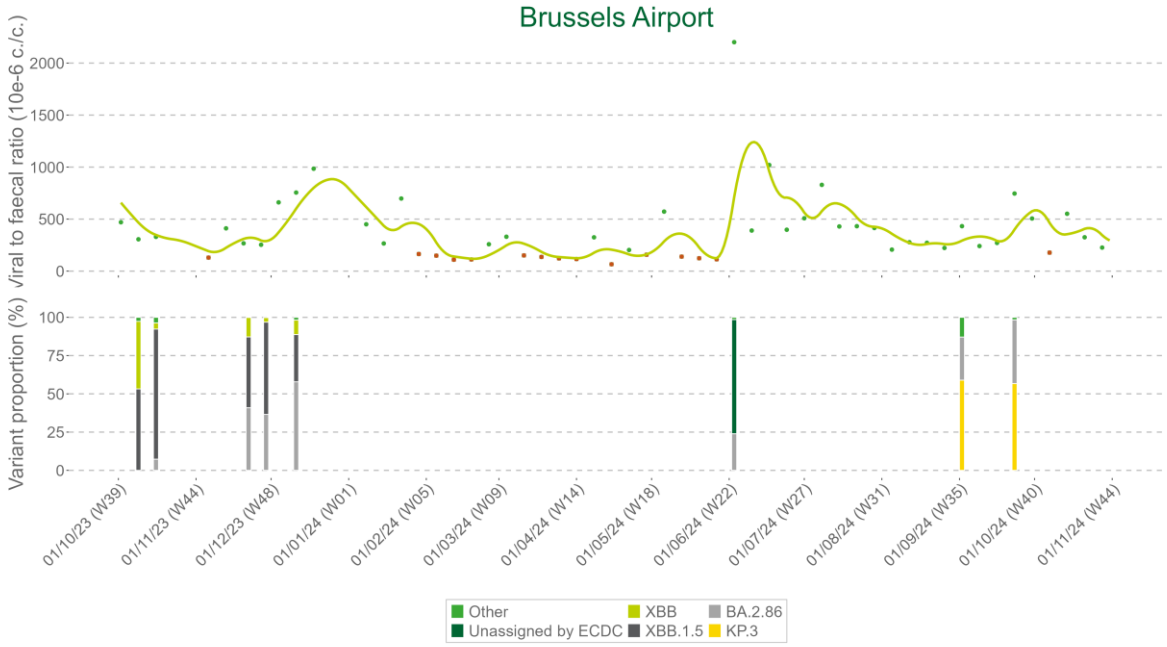


Figure 13 ● Viral ratio expressed as SARS-CoV-2 copies / PMMoV copies (based on the past two weeks moving average) and variant proportion using the ECDC classification for the area of Brussels Airport.

6. ANNEXES

All 30 areas were sampled and are hence included in this week’s report.

Table A.2 ● Variant having a proportion above 10% reported with ECDC and Pango classification for the area of Brussels-North.

Area	Date	ECDC	Pango	Proportion (%)
Brussels-North	2023-10-11	XBB.1.5	EG.5.1	22.8
Brussels-North	2023-10-11	XBB	XBB.2.4	12.7
Brussels-North	2023-10-11	XBB.1.5	XBB.1.16.17	12.1
Brussels-North	2023-10-16	XBB.1.5	EG.5.1.8	17.3
Brussels-North	2023-11-22	BA.2.86	JN.1	26.8
Brussels-North	2023-11-22	BA.2.86	JN.4	13.7
Brussels-North	2023-11-29	BA.2.86	JN.10	16.3
Brussels-North	2023-11-29	BA.2.86	JN.1	15.9
Brussels-North	2023-11-29	BA.2.86	JN.1.1	14.6
Brussels-North	2023-11-29	BA.2.86	JN.1.1.1	11.0
Brussels-North	2023-12-11	BA.2.86	JN.1.1.7	33.7
Brussels-North	2023-12-11	BA.2.86	JN.2.5	26.2
Brussels-North	2023-12-11	BA.2.86	JN.1.22	12.5
Brussels-North	2024-05-27	BA.2.86	JN.1.16	43.5
Brussels-North	2024-05-27	Unassigned by ECDC	XEB	14.5
Brussels-North	2024-05-27	BA.2.86	JN.1.16.3	13.4
Brussels-North	2024-05-27	KP.3	KP.3.1	12.1
Brussels-North	2024-09-02	KP.3	KP.3.1.1	54.8
Brussels-North	2024-09-02	KP.3	MC.16	10.0

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MORE INFORMATION

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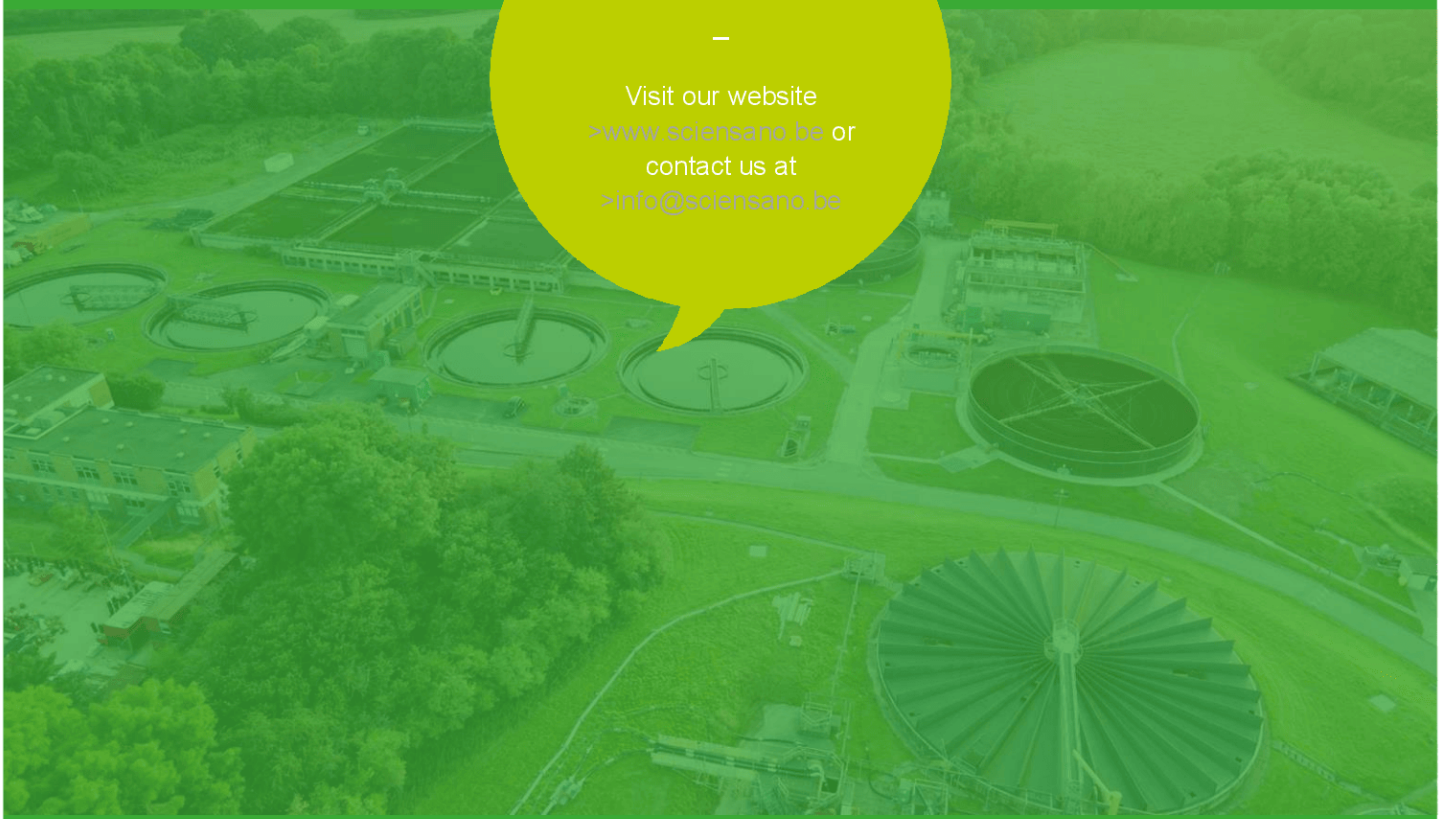

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Responsible editor: C. Léonard, Managing director • Rue Juliette Wytsmanstraat 14 • Brussels • Belgium • D/2024.14.440/28 •
ISSN 2795-7152