

# WEEKLY REPORT – WASTEWATER-BASED EPIDEMIOLOGICAL SURVEILLANCE OF THE SARS-COV-2

## — RESULTS OF 27/04/2022

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In collaboration with:



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# 1. Summary

In the present work, the circulation of the SARS-CoV-2 virus in the environment is assessed based on three indicators. The analysis of wastewater samples collected in 42 wastewater treatment plants covers 45% of the Belgian population, with increased accuracy in the urban areas. The results of the wastewater surveillance are a source of complementary information to the infection cases number. Indeed, the wastewater results do notably include all asymptomatic persons, and are independent of the testing strategy.

Here are the conclusions based on the latest results of April 27<sup>th</sup> 2022:

- At the national level: Since three weeks, the Increasing trend indicator is stable, with approximately one quarter of the areas positive to this indicator. Even though last week, a strong increase in Fast increase has been observed, this week, the Fast increase is at a low level. This has induced a decrease in the High circulation. The decrease in High circulation requires the coming weeks before being confirmed on a longer tendency.
- At the provincial level: All three indicators are not fulfilled in any province. Last week, the Fast increase indicator was fulfilled in 6 provinces. Last week, the Increasing Trend indicator was fulfilled in 1 province. Last week, the High circulation indicator was fulfilled in 4 provinces.
- At the covered areas level: among the 41 areas covered<sup>12</sup>, the number of treatment plants belonging to the different indicators are 3 for the High circulation indicator, 3 for the “Fast increase” indicator and 12 for the “Increasing trend” indicator.
- An alerting situation is evidenced for the covered area of Aartselaar as all three indicators are fulfilled.
- The risk of underestimating the Omicron variant in wastewater was assessed to be negligible.

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The wastewater situation can be followed on a weekly basis on:

- The graphics available on the public [COVID-19 dashboard](#)
- The Risk Assessment Group (RAG) updating the weekly epidemiological situation through a report validated by the Risk Management Group (RMG) and published every Friday in [French](#) and [Dutch](#).
- Further details on the methodology applied for the wastewater surveillance can be found in [the Appendix Methodology document \(access available online\)](#). The methodology was updated on March 29<sup>th</sup> 2022.

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<sup>1</sup> Due to the recent flooding events, the treatment plants from Wegnez (Verviers) and Grosses-Battes (Liège) are temporarily out of order. These two areas had therefore not been screened for SARS-CoV-2. Liège is still well represented within this surveillance, however Verviers is not. Since the 13<sup>th</sup> of October, the covered area of Soumagne is added to the surveillance.

<sup>2</sup> Due to an issue with probes in one of the laboratory, the results for 12 covered areas were unavailable on Monday the 25<sup>th</sup> of April. This concerns the areas of Aartselaar, Antwerpen-Zuid, Antwerpen-Noord, Dendermonde, Deurne, Genk, Hasselt, Houthalen-Centrum, Mechelen-Noord, Sint-Niklaas, Tessenderlo and Turnhout. This absence of samples was estimated to have minor impact on the indicators.

## 2. Introduction

In mid-September 2020 started the SARS-CoV-2 national wastewater surveillance project. The present report is an outcome of this project aiming to assess weekly the wastewater-based epidemiological situation of Belgium.

The surveillance is based on the analysis of water samples collected twice per week from the influent of 42 WasteWater Treatment Plants (WWTPs) spread over Belgium. The evolution of the SARS-CoV-2 wastewater concentrations measured over time in the different treatment plants is analysed at different levels: regional, provincial, and the catchment area covered by the individual treatment plants. Also, three alerting indicators were developed to highlight areas of concern regarding the high circulation, the fast evolution, and the increasing trend of the observed wastewater concentrations.

In this report, the weekly wastewater-based epidemiological situation is presented and discussed at the above-mentioned levels based on the three categories of alerting indicators. Moreover, the remaining sources of uncertainties are discussed together with their expected impacts on the wastewater results interpretation.

## 3. Methodology

### 3.1. SAMPLE COLLECTION AND ANALYSIS

The surveillance project, which started in mid-September 2020 covers around 45% of the Belgian population. The population covered is 40% in the Flemish region, 35% in the Walloon region, and nearly 100% in the Brussels region. Figure 1 shows the catchment areas covered by the 42 WWTPs included in the project, which are located in high population density areas. A catchment area is defined by the area delimiting the population covered by a specific wastewater sample, collected at the inlet of the WWTPs. Further coverage details can be found in Table 2 by province (see also Table A1 in [the Appendix Methodology document \(access available online\)](#) ) and on the [Sciensano public dashboard](#). In practice, 24-hour composite samples are collected twice a week on Mondays and Wednesdays from the influent of WWTP and are analysed for the presence of SARS-CoV-2 RNA by three different laboratories. The resulting SARS-CoV-2 concentrations (3 targeted genes) are delivered to Sciensano within 2 days for data analysis purpose. Concretely, the results which are representative of Mondays and Wednesdays are respectively available on Wednesdays and Fridays.

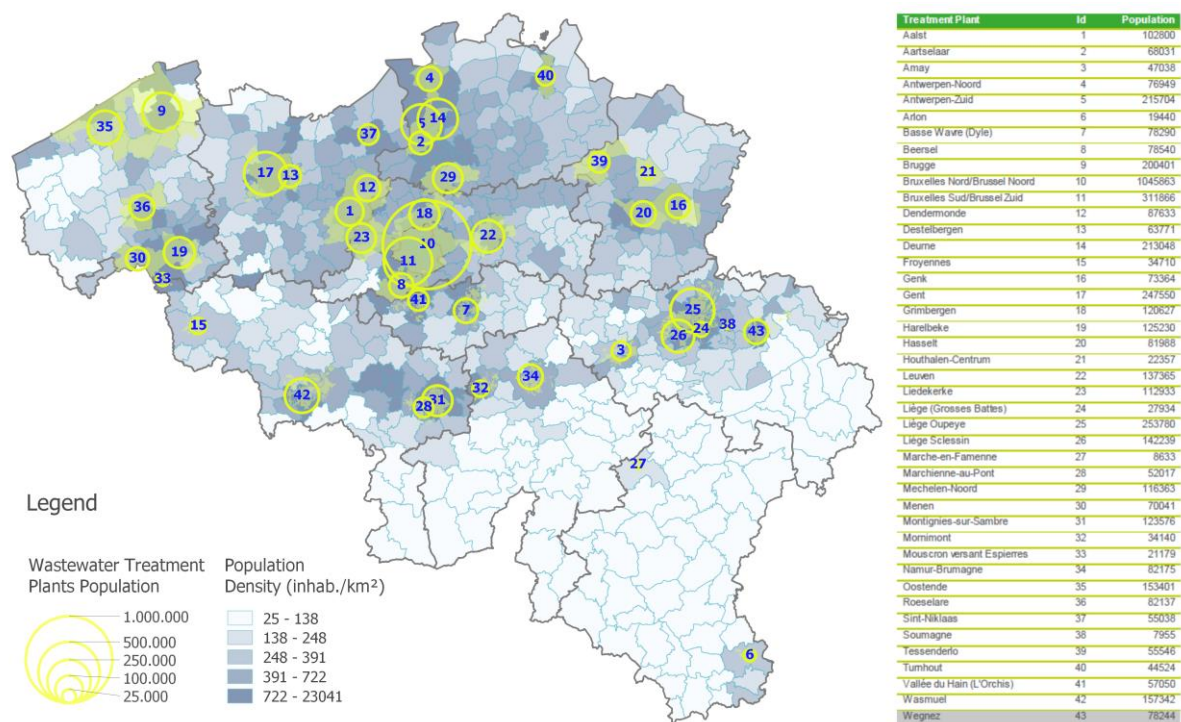


Figure 1: The population located in the areas covered by the wastewater treatment plants (highlighted in yellow) and the population density for each municipality (indicated by the blue scale). Note that due to the recent flooding, the treatment plants of Wegnez and Liège (Grosses Battes) are out of order. These areas are, hence, not considered anymore in the wastewater surveillance and has been partially replaced by the treatment plant of Soumagne.

### 3.2. WASTEWATER RESULTS

The quantification of SARS-CoV-2 in RNA copies/ ml (concentration) was generalized in mid-February 2021 to all the treatment plants investigated. Before this date, the quantitative values were estimated based on the retrospective application of the quantification method (see details in Appendix) from mid-September 2020 to mid-February 2021 for Flanders and Brussels. In Wallonia, the quantitative results were available since the start.

Preliminary results allowed for estimating the limit of quantification of the analytical method at 20 copies/ml.

In order to account for possible dilution by rainy events and for the number of people persons living in the catchment of each WWTP, a correction is applied on the viral concentration: the concentration is divided by the inlet flow and by the number of inhabitant equivalent of its respective WWTP and expressed by 100k inhabitant. This correction changes the reporting units from copy/ml into copy/day/100k inhabitants equivalent. We will refer this new signal as viral load instead of viral concentration.

### 3.3. ALERTING INDICATORS

To highlight the areas of possible concern, the three following types of alerting indicators are assessed twice a week, based on the viral load (RNA copies/day/100k inhabitants) measured for the three targeted gene fragments (E, N1, and N2):

1. The **High circulation** indicator highlights the catchment areas where the viral loads are high. It corresponds to a situation where the viral loads exceed half of the highest value recorded during the fifth wave (i.e. from first of January 2022 till first of March 2022).

2. The **Fast increase** indicator highlights the catchment areas where the viral loads have rapidly increased for the last week. It corresponds to a situation where the moving average on the past 7 days of the viral load has increased faster than 70% per week if being above the estimated limit of quantification. The increasing slope is normalized for each treatment plant.
3. The **Increasing trend** indicator highlights the catchment areas where the viral loads have been increasing for more than 6 days. The indicator is computed based on the moving average on the past two weeks of the viral load.

The indicators were developed in order to be able to track the different phases of an outbreak. Typically, when the viral loads in wastewaters in an area start to increase, the indicator Increasing trend will be the first fulfilled. If the viral loads increase quickly, the Fast increase indicator will be fulfilled. Finally, after the initial increasing phase, the viral loads in an area may be sufficiently high to result in the High circulation to be fulfilled.

It should be noted that in several areas, the viral load exceeded the maximal viral load registered during the 3rd wave. The date at which the maximal viral load has been reached for each area is indicated in the tables A1 to A4 in the appendices.

### 3.4. CASES RESULTS

The cases number data presented in this report come from the COVID-19 laboratory-confirmed cases database centralized by Sciensano. The cases number, used to compute the 14 days-incidence, only accounts for the physical areas covered by the wastewater project (see Figure 1).

### 3.5. CAUTION POINTS FOR THE RESULTS INTERPRETATION

Only the trends, observed through the alerting indicators, should be assessed for the comparison of different areas. As the viral load values differ from a WWTP to another, notably due to the differences in population sizes covered, absolute values should not be compared. Additionally, the situation comparison between the regions should be considered with caution. The degree of comparability is not yet known and depends on the comparability of the results between the different laboratories performing the analyses (see [the Appendix Methodology document \(access online\)](#)).

Moreover, the wastewater viral loads and the cases numbers presented in this report do not originate from the same population, even though the positive cases are selected only for the municipalities covered by the wastewater surveillance. For instance, the wastewater results account for all infected persons (whatever age or symptomatic condition) while the cases numbers include only the persons clinically tested positive. Likewise, an infected person covered by the wastewater results could be associated with another area in the clinical surveillance as the person's postal code is used for clinical statistics, i.e. mobility bias. Therefore, the correlation between the wastewater viral load and the cases number varies according to the area considered. The wastewater results are thus complementary and independent information to the results of the cases.

The correlation between the wastewater viral loads and the cases numbers could also be influenced by the vaccination campaign and the circulation of variants.

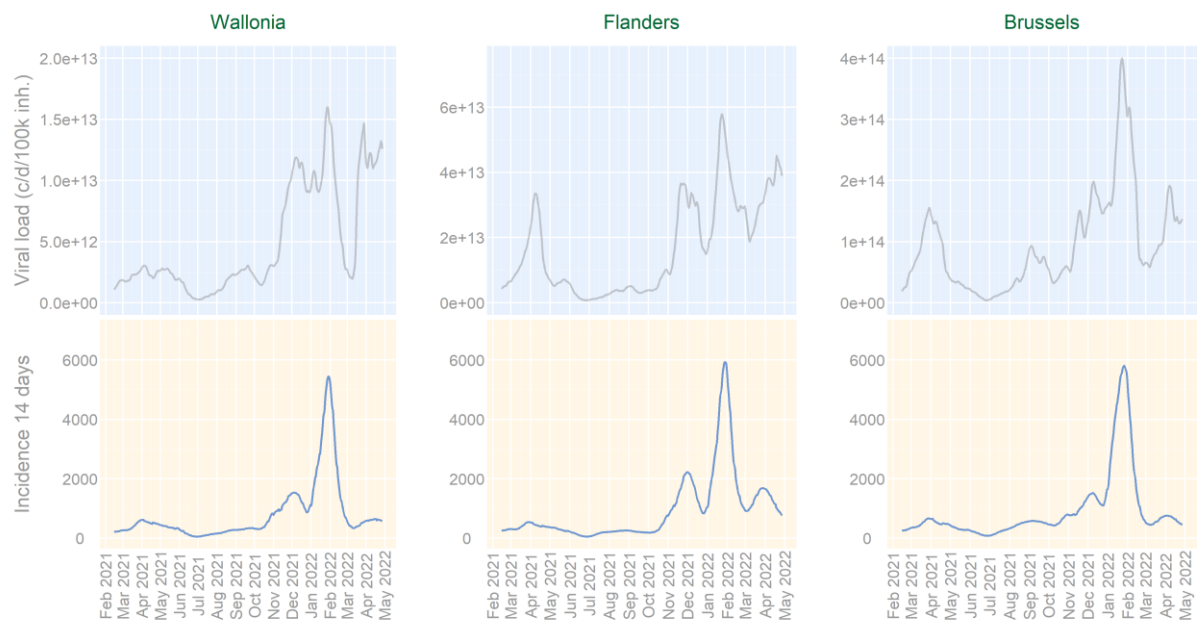
## 4. Results

### 4.1. REGIONS

Figure 2 represents the quantitative SARS-CoV-2 RNA viral loads in the wastewaters and the 14 days incidence for each region, compiling the incidence data of the area covered by the wastewater surveillance.

Several waves can be seen in Figure 2:

- The 3rd wave occurring in March 2021.
- The 4th wave occurring in December 2021.
- The 5th wave occurring in January 2022.



**Figure 2: The SARS-CoV-2 RNA viral loads expressed as copies/days/100k habitants (based on the past two weeks moving average applied on the linear interpolation) (Top), and the 14 days incidence in the population covered by the wastewater surveillance since mid-February 2021 (bottom).**

Table 1 shows, for each region, the results associated with the samples of Wednesday January 26th 2022.

Table 1 allows to track the changes between the situation as of today (April 27<sup>th</sup> 2022) and the situation as of last week (April 20<sup>th</sup> 2022). Hereby, two distinct cases are taken into account:

1. If a region has at least one indicator fulfilled this week and it was not the case last week, its **name is displayed in bold** in the table.
2. If a region has at least one indicator fulfilled this week and if it also was the case last week, any change in indicator fulfilment (i.e. if the value for any indicator has changed from 0 to 1 or from 1 to 0) is indicated in **coloured bold text**.

The last trends were analysed based on the alerting indicators presented in Table 1. All three indicators are not fulfilled all three regions. Hence, the rapid increase observed in the Fast increase indicator identified last week has changed and the viral loads are now decreasing in all three regions. This has led to the high circulation in Wallonia not to be fulfilled anymore this week.

Important to remember that the High circulation is highlighted when the viral circulation level has reached half of the highest level of viral load obtained during the 5th wave. As this maximum is much higher for the 5th wave than the 3rd wave, which was our reference before 2022 for the High circulation indicator, not having the High circulation level highlighted does not mean that the virus is not still circulating a lot in comparison to the 3rd wave.

**Table 1: Alerting indicators fulfilled (1) or not (0) on April 27<sup>th</sup> 2022. Columns represent the population coverage of the regions (Pop. coverage) and the three alerting indicators High Circulation (High), Fast increase (Fast) and Increasing trend (Incr.). The specifications of the four last columns are explained in the footnotes 1-4 below the table. Missing data is indicated with a “/”.**

Region	Pop. coverage	High	Fast	Incr.	Norm. viral load (%) <sup>1</sup>	Mean viral load <sup>2</sup>	Norm. evol. (%/week) <sup>3</sup>	Incr. days <sup>4</sup>
Brussels	100%	0	<b>0</b>	0	12	73	-62	5
Flanders	41%	0	<b>0</b>	0	18	16	-38	2
Wallonia	31%	<b>0</b>	<b>0</b>	0	24	6.2	-9	2

<sup>1</sup> : the viral load normalized with the maximum viral load measured in the corresponding catchment area during the fifth wave (i.e. from first of January 2022 till the first of march 2022).

<sup>2</sup> : the viral load computed on the mean of the replicate of the three targeted gene fragments as explained in section “3.2 Wastewater results”. The mean viral load is expressed in 10<sup>12</sup> copies/day/100k inhabitants.

<sup>3</sup> : the slope (%/week) of the past 7 days moving average of the viral load if the corresponding concentration is above the estimated limit of quantification.

<sup>4</sup> : the cumulative number of days of increase of the past 14 days moving average of the viral load.

## 4.2. PROVINCES

Table 2 shows, for each Province, the results associated with the samples of Wednesday April 27<sup>th</sup> 2022.

Table 2 allows to track the changes between the situation as of today (April 27<sup>th</sup> 2022) and the situation as of last week (April 20<sup>th</sup> 2022). Hereby, two distinct cases are taken into account:

1. If a province has at least one indicator fulfilled this week and it was not the case last week, its **name is displayed in bold** in the table.
2. If a province has at least one indicator fulfilled this week and if it also was the case last week, any change in indicator fulfilment (i.e. if the value for any indicator has changed from 0 to 1 or from 1 to 0) is indicated in **coloured bold text**.

Table 2 shows, for each Province, the results associated with the samples of Wednesday April 27<sup>th</sup> 2022, for the three alerting indicators:

- The High circulation indicator is not fulfilled in any province. Last week, this indicator was fulfilled in 4 provinces.
- The Fast increase indicator is not fulfilled in any province. Last week, this indicator was fulfilled in 6 provinces.
- The Increasing trend indicator is not fulfilled in any province. Last week, this indicator was fulfilled in 1 province.
- It should be noted that although the 50% threshold of viral circulation is not exceeded (in comparison to the highest level observed during the 5th wave), the viral load in the province of Hainaut remains high (with 43%).



**Table 2: Alerting indicators fulfilled (1) or not (0) on April 27<sup>th</sup> 2022. Columns represent the population coverage of the WWTPs within the Province (Pop. coverage) and the three alerting indicators High circulation (High), Fast increase (Fast) and Increasing trend (Incr.). The specifications of the four last columns are explained in the footnotes 1-4 below the table. Missing data is indicated with a “/”.**

Province	Pop. coverage	High	Fast	Incr.	Norm. viral load (%) <sup>1</sup>	Mean viral load <sup>2</sup>	Norm. evol. (%/w) <sup>3</sup>	Incr. days <sup>4</sup>
Antwerpen	41%	0	0	0	13	4.3	-4	5
Brabant Wallon	33%	0	0	0	38	7.5	-6	1
Brussels	100%	0	0	0	12	73	-62	5
Hainaut	29%	0	0	0	43	11	-11	5
Liège	50%	0	0	0	4	2.3	-23	0
Limburg	26%	0	0	0	18	7.1	-27	5
Luxembourg	10%	0	0	0	15	8.7	-17	0
Namur	23%	0	0	0	24	4.4	47	2
Oost-Vlaanderen	38%	0	0	0	17	15	-25	2
Vlaams-Brabant	61%	0	0	0	17	27	-76	0
West-Vlaanderen	52%	0	0	0	25	26	-64	0

<sup>1</sup> : the viral load normalized with the maximum viral load measured in the corresponding catchment area during the fifth wave (i.e. from first of January 2022 till the first of march 2022).

<sup>2</sup> : the viral load computed on the mean of the replicates of the three targeted gene fragments as explained in section “3.2 Wastewater results”. The mean viral load is expressed in 10<sup>12</sup> copies/day/100k inhabitants.

<sup>3</sup> : the slope (%/week) of the past 7 days moving average of the viral load if the corresponding concentration is above the estimated limit of quantification.

<sup>4</sup> : the cumulative number of days of increase of the past 14 days moving average of the viral load.

### 4.3. CATCHMENT AREAS

Table 3 shows, for each catchment area, the values of the three alerting indicators obtained based on the results of last Wednesday’s sample. Table 3 is a snapshot of the number of areas highlighted by the indicators for the last results obtained.

Table 3 allows to track the changes between the situation as of today (April 27<sup>th</sup> 2022) and the situation as of last week (April 20<sup>th</sup> 2022). Hereby, three distinct cases are taken into account:

1. If an area has at least one indicator fulfilled this week and it was not the case last week, its **name is displayed in bold** in the table.
2. If an area has at least one indicator fulfilled this week and if it also was the case last week, any change in indicator fulfilment (i.e. if the value for any indicator has changed from 0 to 1 or from 1 to 0) is indicated in **coloured bold text**.
3. Any area which had at least one indicator fulfilled last week but not this week is listed below Table 3.

Here are the results associated with the samples of April 27<sup>th</sup> 2022:

- The High circulation indicator is fulfilled in 3 covered areas: Mouscron versant Espierres (109%), Vallée du Hain (L'Orchis) ( 52%) and Aartselaar ( 51%) (see Appendix A1 for more details). Last week, this indicator was fulfilled in 10 covered areas.
- The Fast increase is fulfilled in 3 covered areas: Aartselaar (313% increase per week), Mornimont (142% increase per week) and Vallée du Hain (L'Orchis) (82% increase per week) (see Appendix A2 for more details). Last week, this indicator was fulfilled in 14 covered areas.
- The Increasing trend indicator is fulfilled in 12 covered areas. Amongst these areas, the viral load is continually increasing since more than two weeks in 2 areas: Marchienne-au-Pont (14 days) and Mechelen-Noord (14 days). Further details can be found in Appendix A3. Last week, this indicator was fulfilled in 10 covered areas.
- An alerting situation is evidenced for the covered area of Aartselaar as all three indicators are fulfilled.

The wastewater results at the level of the local covered areas can be accessed online for each area on the [COVID-19 dashboard](#).

**Table 3: Alerting indicators fulfilled (1) or not (0) on April 27<sup>th</sup> 2022. Columns represent the provinces, different WWTPs within the Provinces and the three alerting indicators High circulation (High), Fast increase (Fast) and Increasing trend (Incr.). The specifications of the four last columns are explained in the footnotes 1-4 below the table. Missing data is indicated with a “/”.**

Province	WWTP	High	Fast	Incr.	Norm. viral load (%) <sup>1</sup>	Mean viral load <sup>2</sup>	Norm evol. (%/week) <sup>3</sup>	Incr. days <sup>4</sup>
Antwerpen	Aartselaar	1	1	1	51	11	313	9
Brussels	Brussels-North	0	0	1	12	79	-68	7
Oost-Vlaanderen	<b>Dendermonde</b>	0	0	1	14	3.3	38	7
Antwerpen	<b>Deurne</b>	0	0	1	5	3.5	-66	7
Limburg	<b>Genk</b>	0	0	1	26	11	23	7
Limburg	<b>Hasselt</b>	0	0	1	17	7.9	-76	7
Hainaut	Marchienne-au-Pont	0	0	1	44	15	1	14
Antwerpen	Mechelen-Noord	0	0	1	17	5.3	52	14
Hainaut	Montignies-sur-Sambre	0	0	1	44	11	23	7
Namur	<b>Mornimont</b>	0	1	1	23	5.6	142	7
Hainaut	Mouscron versant Espierres	1	0	1	109	53	32	9
Oost-Vlaanderen	<b>Sint-Niklaas</b>	0	0	1	10	6	36	7
Brabant Wallon	<b>Vallée du Hain (L'Orchis)</b>	1	1	0	52	7.8	82	0

<sup>1</sup> : the viral load normalized with the maximum viral load measured in the corresponding catchment area during the fifth wave (i.e. from first of January 2022 till the first of march 2022).

<sup>2</sup> : the viral load computed on the mean of the replicates of the three targeted gene fragments as explained in section “3.2 Wastewater results”; The mean viral load is expressed in 10<sup>12</sup> copies/day/100k inhabitants.

<sup>3</sup> : the slope (%/week) of the past 7 days moving average of the viral load if the corresponding concentration is above the estimated limit of quantification.

<sup>4</sup> : the cumulative number of days of increase of the past 14 days moving average of the viral load.

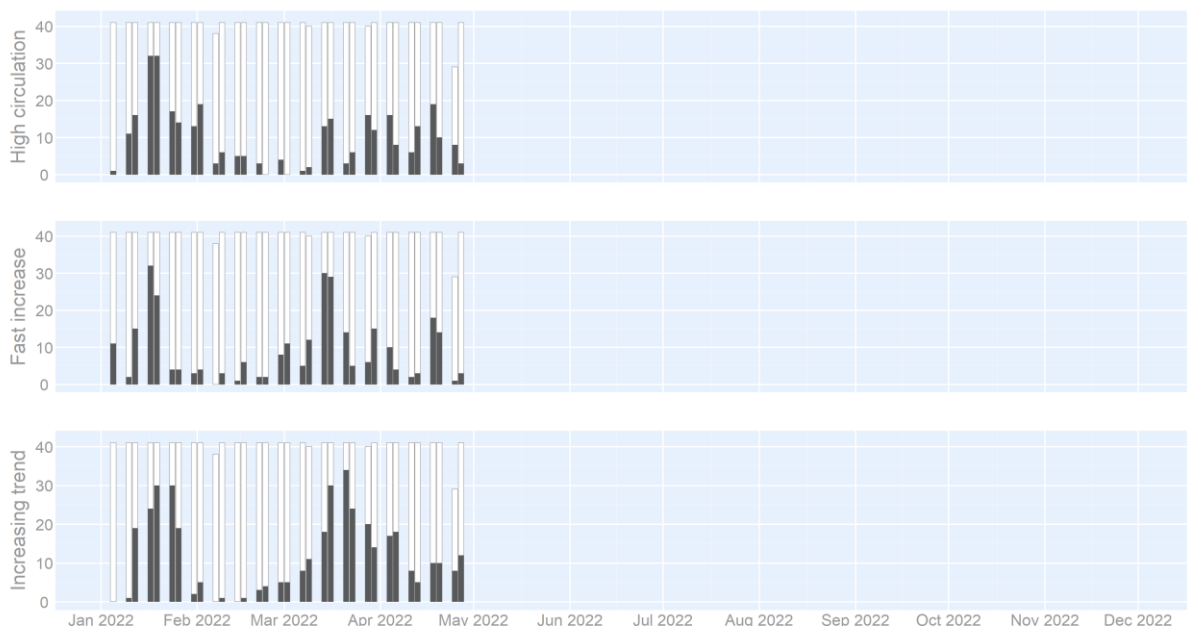
The following areas had the corresponding indicator fulfilled last week but not this week:

- High circulation indicator: Aalst, Froyennes, Liège Oupeye, Marche-en-Famenne, Menen, Namur-Brumagne and Wasmuel.
- Fast increase indicator: Aalst, Basse Wavre (Dyle), Brugge, Destelbergen, Grimbergen, Harelbeke, Leuven, Liedekerke, Liège Oupeye and Menen.
- Increasing trend indicator: Aalst, Antwerpen-North, Destelbergen, Froyennes, Harelbeke, Liège Oupeye, Liège Sclessin and Wasmuel.

Further details on covered area without fulfilled indicators can be found in Table A4.

Figure 3 was developed to offer a dynamic view of the three alerting indicators over time. For further insights on the dynamic of the different indicators, see Section 3.3.

Since three weeks, the Increasing trend indicator is stable, with approximately one quarter of the areas positive to this indicator. Even though last week, a strong increase in Fast increase has been observed, this week, the Fast increase is at a low level. This has induced a decrease in the High circulation. The decrease in High circulation requires the coming weeks before being confirmed on a longer tendency.



**Figure 3: The number of areas (among the 41 covered by the wastewater surveillance this week and the 42 normally considered), with positive alerting indicators (black) compared to the total number of areas considered every week in the wastewater surveillance (white). Latest results (last bar) are from April 27<sup>th</sup> 2022. The white bars represent the total number of areas considered every week in the wastewater surveillance.**

#### 4.4. VARIANTS

The Omicron variant represents a high proportion of all the reported cases in Belgium<sup>3</sup>. The impact of the Omicron variant was assessed to be negligible for the following reason: mutations present in the Omicron variant on the N1 and E genes could have potentially led to a drop out of those two genes leading to an underestimation of the viral loads in wastewaters. Preliminary experimental lab results indicate that no drop out of the genes should be expected. Hence, the sensitivity of the analytical lab methods used was not impacted. Therefore, the circulation of the Omicron variant is not underestimated in wastewaters.

However, the Omicron variant might have an impact on the faecal shedding load if infected people would excrete less in their faeces than for Delta. That stays hypothetical as we do not have strong evidence on it yet.

<sup>3</sup> Source :[Molecular surveillance of SARS-CoV-2](#)

A regular investigation of the possible impact of variants of concern is done in order to screen their evolution in time.

## 5. Appendix – Areas classified by alerting indicator

**A 1: Covered areas (3 out of 41 on April 27<sup>th</sup> 2022) characterized as High circulation sorted in the descending order of importance.**

Province	WWTP	High	Fast	Incr.	Norm. viral load (%) <sup>1</sup>	Mean viral load <sup>2</sup>	Norm evol. (%/week) <sup>3</sup>	Incr. days <sup>4</sup>	Date Max cc <sup>5</sup>
Hainaut	Mouscron versant Espierres	1	0	1	109	53	32	9	27/04/2022
Brabant Wallon	Vallée du Hain (L'Orchis)	1	1	0	52	7.8	82	0	5 <sup>th</sup> wave
Antwerpen	Aartselaar	1	1	1	51	11	313	9	20/04/2022

<sup>1</sup> : the viral load normalized with the maximum viral load measured in the corresponding catchment area during the fifth wave (i.e. from first of January 2022 till the first of March 2022).

<sup>2</sup> : the viral load computed on the mean of the replicates of the three targeted gene fragments as explained in section "3.2 Wastewater results". The mean viral load is expressed in 10<sup>12</sup> copies/day/100k inhabitants.

<sup>3</sup> : the slope (%/week) of the past 7 days moving average of the viral load if the corresponding concentration is above the estimated limit of quantification.

<sup>4</sup> : the cumulative number of days of increase of the past 14 days moving average of the viral load.

<sup>5</sup> : date at which the measured viral load was the highest since the beginning of 2022. If the date was between the first of January 2022 and the first of March 2022, the date is considered to be during the fifth wave and mentioned as such.

**A 2: Covered areas (3 out of 41 on the April 27<sup>th</sup> 2022) characterized as Fast increase sorted in the descending order of importance.**

Province	WWTP	High	Fast	Incr.	Norm. viral load (%) <sup>1</sup>	Mean viral load <sup>2</sup>	Norm evol. (%/week) <sup>3</sup>	Incr. days <sup>4</sup>	Date Max cc <sup>5</sup>
Antwerpen	Aartselaar	1	1	1	51	11	313	9	20/04/2022
Namur	Mornimont	0	1	1	23	5.6	142	7	5 <sup>th</sup> wave
Brabant Wallon	Vallée du Hain (L'Orchis)	1	1	0	52	7.8	82	0	5 <sup>th</sup> wave

<sup>1</sup> : the viral load normalized with the maximum viral load measured in the corresponding catchment area during the fifth wave (i.e. from first of January 2022 till the first of March 2022).

<sup>2</sup> : the viral load computed on the mean of the replicates of the three targeted gene fragments as explained in section "3.2 Wastewater results". The mean viral load is expressed in 10<sup>12</sup> copies/day/100k inhabitants.

<sup>3</sup> : the slope (%/week) of the past 7 days moving average of the viral load if the corresponding concentration is above the estimated limit of quantification.

<sup>4</sup> : the cumulative number of days of increase of the past 14 days moving average of the viral load.

<sup>5</sup> : date at which the measured viral load was the highest since the beginning 2022. If the date was between the first of January 2022 and the first of March 2022, the date is considered to be during the fifth wave and mentioned as such.

**A 3: Covered areas (12 out of 41 on the April 27<sup>th</sup> 2022) characterized as Increasing trend sorted in the descending order of importance.**

Province	WWTP	High	Fast	Incr.	Norm. viral load (%) <sup>1</sup>	Mean viral load <sup>2</sup>	Norm evol. (%/week) <sup>3</sup>	Incr. days <sup>4</sup>	Date Max cc <sup>5</sup>
Hainaut	Marchienne-au-Pont	0	0	1	44	15	1	14	5 <sup>th</sup> wave
Antwerpen	Mechelen-Noord	0	0	1	17	5.3	52	14	20/04/2022
Antwerpen	Aartselaar	1	1	1	51	11	313	9	20/04/2022
Hainaut	Mouscron versant Espierres	1	0	1	109	53	32	9	27/04/2022
Brussels	Brussels-North	0	0	1	12	79	-68	7	5 <sup>th</sup> wave
Oost-Vlaanderen	Dendermonde	0	0	1	14	3.3	38	7	5 <sup>th</sup> wave
Antwerpen	Deurne	0	0	1	5	3.5	-66	7	5 <sup>th</sup> wave
Limburg	Genk	0	0	1	26	11	23	7	09/03/2022
Limburg	Hasselt	0	0	1	17	7.9	-76	7	5 <sup>th</sup> wave
Hainaut	Montignies-sur-Sambre	0	0	1	44	11	23	7	5 <sup>th</sup> wave
Namur	Mornimont	0	1	1	23	5.6	142	7	5 <sup>th</sup> wave
Oost-Vlaanderen	Sint-Niklaas	0	0	1	10	6	36	7	5 <sup>th</sup> wave

<sup>1</sup> : the viral load normalized with the maximum viral load measured in the corresponding catchment area during the fifth wave (i.e. from first of January 2022 till the first of March 2022).

<sup>2</sup> : the viral load computed on the mean of the replicates of the three targeted gene fragments as explained in section "3.2 Wastewater results". The mean viral load is expressed in 10<sup>12</sup> copies/day/100k inhabitants.

<sup>3</sup> : the slope (%/week) of the past 7 days moving average of the viral load if the corresponding concentration is above the estimated limit of quantification.

<sup>4</sup> : the cumulative number of days of increase of the past 14 days moving average of the viral load.

<sup>5</sup> : date at which the measured viral load was the highest since the beginning of 2022. If the date was between the first of January 2022 and the first of March 2022, the date is considered to be during the fifth wave and mentioned as such.

#### A 4: Covered areas (28 out of 41 on the April 27<sup>th</sup> 2022) in which no alerting indicator are fulfilled.

Province	WWTP	High	Fast	Incr.	Norm. viral load (%) <sup>1</sup>	Mean viral load <sup>2</sup>	Norm evol. (%/week) <sup>3</sup>	Incr. days <sup>4</sup>	Date Max cc <sup>5</sup>
Oost-Vlaanderen	Aalst	0	0	0	18	11	-74	0	18/04/2022
Liège	Amay	0	0	0	18	6	-46	0	5 <sup>th</sup> wave
Antwerpen	Antwerpen-North	0	0	0	14	3.1	20	0	5 <sup>th</sup> wave
Antwerpen	Antwerpen-South	0	0	0	7	3.1	-69	0	5 <sup>th</sup> wave
Luxembourg	Arlon	0	0	0	15	11	3	0	5 <sup>th</sup> wave
Brabant Wallon	Basse Wavre (Dyle)	0	0	0	28	7.2	-70	2	16/03/2022
Vlaams-Brabant	Beersel	0	0	0	12	32	-70	0	5 <sup>th</sup> wave
West-Vlaanderen	Brugge	0	0	0	15	9.5	-74	0	18/04/2022
Brussels	Brussel-South	0	0	0	11	53	-45	0	5 <sup>th</sup> wave
Oost-Vlaanderen	Destelbergen	0	0	0	12	17	-81	0	18/04/2022

Province	WWTP	High	Fast	Incr.	Norm. viral load (%) <sup>1</sup>	Mean viral load <sup>2</sup>	Norm. evol. (%/week) <sup>3</sup>	Incr. days <sup>4</sup>	Date Max cc <sup>5</sup>
Hainaut	Froyennes	0	0	0	23	3.2	-57	0	16/03/2022
Oost-Vlaanderen	Gent	0	0	0	21	22	-26	0	5 <sup>th</sup> wave
Vlaams-Brabant	Grimbergen	0	0	0	10	24	-73	0	5 <sup>th</sup> wave
West-Vlaanderen	Harelbeke	0	0	0	19	27	-70	2	5 <sup>th</sup> wave
Limburg	Houthalen-Centrum	0	0	0	27	3.2	63	0	5 <sup>th</sup> wave
Vlaams-Brabant	Leuven	0	0	0	13	24	-81	0	18/04/2022
Vlaams-Brabant	Liedekerke	0	0	0	34	30	-77	0	18/04/2022
Liège	Liège Oupeye	0	0	0	0	0	0	0	28/03/2022
Liège	Liège Sclessin	0	0	0	7	4.8	-55	0	16/03/2022
Luxembourg	Marche-en-Famenne	0	0	0	15	4.4	-61	0	23/03/2022
West-Vlaanderen	Menen	0	0	0	14	22	-75	0	18/04/2022
Namur	Namur-Brumagne	0	0	0	24	3.9	8	0	5 <sup>th</sup> wave
West-Vlaanderen	Oostende	0	0	0	43	45	-52	0	04/04/2022
West-Vlaanderen	Roeselare	0	0	0	37	33	-46	0	18/04/2022
Liège	Soumagne	0	0	0	15	12	-32	0	28/03/2022
Limburg	Tessenderlo	0	0	0	7	2.6	-58	0	5 <sup>th</sup> wave
Antwerpen	Turnhout	0	0	0	8	3.6	-73	0	5 <sup>th</sup> wave
Hainaut	Wasmuel	0	0	0	39	4.8	-38	0	16/03/2022

<sup>1</sup> : the viral load normalized with the maximum viral load measured in the corresponding catchment area during the fifth wave (i.e. from first of January 2022 till the first of March 2022).

<sup>2</sup> : the viral load computed on the mean of the replicates of the three targeted gene fragments as explained in section "3.2 Wastewater results". The mean viral load is expressed in 10<sup>12</sup> copies/day/100k inhabitants.

<sup>3</sup> : the slope (%/week) of the past 7 days moving average of the viral load if the corresponding concentration is above the estimated limit of quantification.

<sup>4</sup> : the cumulative number of days of increase of the past 14 days moving average of the viral load.

<sup>5</sup> : date at which the measured viral load was the highest since the beginning of 2022. If the date was between the first of January 2022 and the first of March 2022, the date is considered to be during the fifth wave and mentioned as such.