STATENS SERUM INSTITUT

Methodology behind the published SARS-CoV-2 data from wastewater surveillance in Denmark

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1: Purpose of the method description

The purpose of this document is to provide the reader with insights into how surveillance of SARS-CoV-2 in wastewater is carried out in Denmark from sampling to sharing the data via our website (Figure 1). Furthermore, this document explains the data processing and analyses behind the data available for download on the website.

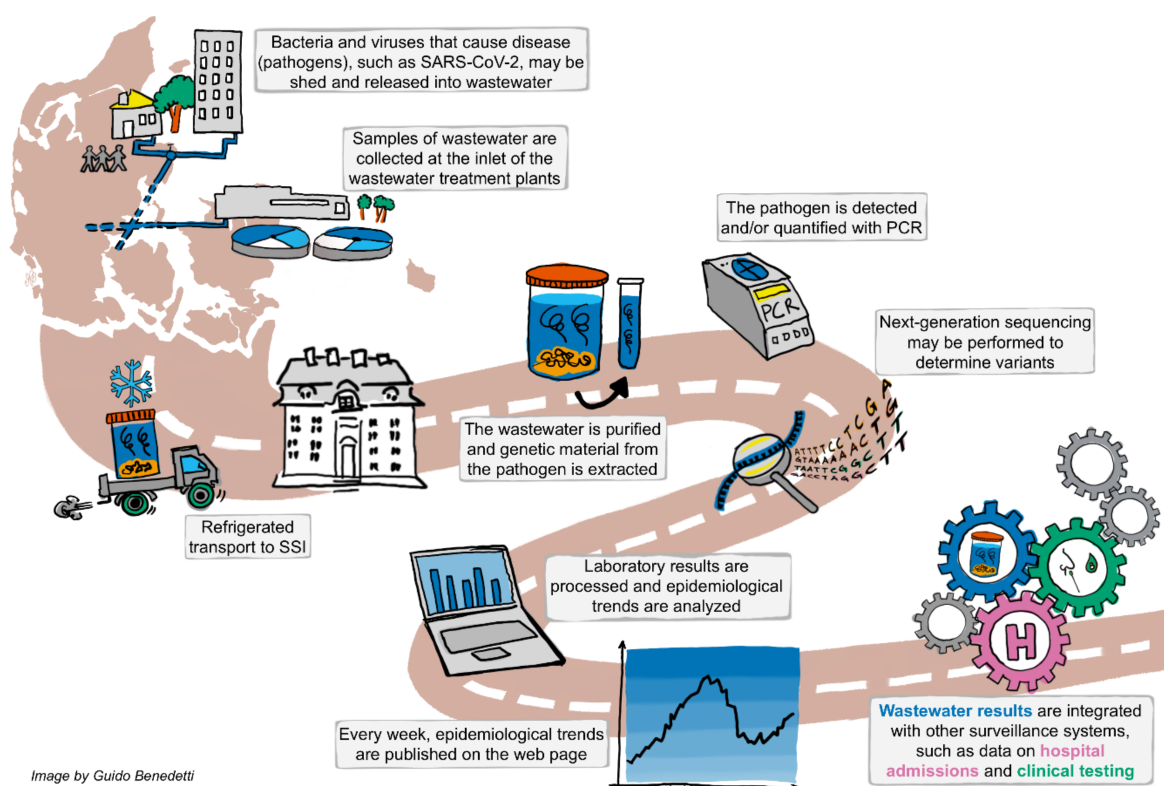

Figure 1: Schematic overview over the pathogen wastewater surveillance in Denmark.

2: How is SARS-CoV-2 measured in wastewater?

Detection of SARS-CoV-2 virus in the wastewater starts with collection of samples at the inlet of wastewater treatment plants. The samples are then transported cold to Statens Serum Institut (SSI), where RNA extraction and reverse transcription quantitative polymerase chain reaction (RT-qPCR) takes place in the laboratory to measure the quantity of SARS-CoV-2 virus in the wastewater samples.

For the national wastewater surveillance of SARS-CoV-2, 29 wastewater samples are currently collected from 28 wastewater treatment plants across Denmark; two samples are taken from two separate inlets of one treatment plant in Copenhagen (Lynetten).

2.1 Current sampling

- 29 inlet samples are taken from 28 treatment plants once a week
- 24 wastewater treatment plants collect 24-hour flow-proportional composite samples with autosamplers
- Four wastewater treatment plants collect 24-hour time-proportional composite samples with autosamplers
- Samples are transported and stored at 4°C and analyzed within 24 hours of collection

2.2 Coverage of wastewater surveillance

Since surveillance of SARS-CoV-2 in wastewater was established, the magnitude of coverage has varied due to changing priorities and resources. If the dataset for download is used, it is important to take the sampling intensity and coverage into account. A change log accompanies the dataset when downloaded.

- From 01.02.2026 and onwards, 29 samples are collected from 28 wastewater treatment plants once weekly (Figure 2)
- Between 04.02.2023 and 31.01.2026, 29 samples were taken from 28 wastewater treatment plants twice a week
- In the period from 09.07.2022 to 03.02.2023, 87 samples were taken from 83 wastewater treatment plants twice a week. In addition, two weekly samples from seven decentralized sampling sites were included.
- In the period from 01.01.2022 to 08.07.2022, 202 samples were taken from 198 treatment plants and 26 samples from decentralized sampling sites three times a week.

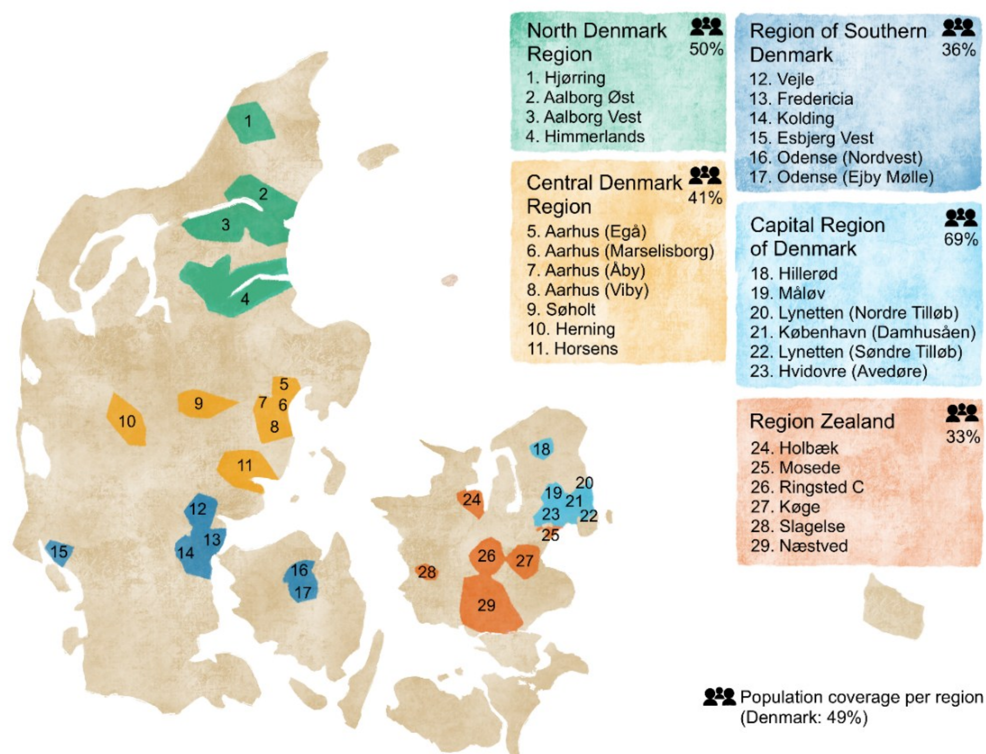

Figure 2: Overview over currently included wastewater catchment areas, where pathogen surveillance in wastewater is performed.

2.3 Brief outline of laboratory procedures

The laboratory analysis of wastewater samples from Denmark has been performed at SSI since 01.04.2023*. Firstly, virus particles are concentrated from 10 ml wastewater sample using the NanoTrap® Microbiome A particles (Ceres Nanosciences). These Microbiome A particles are magnetic and made of a highly porous hydrogel polymer with affinity capture, that allow for selection and concentration of microorganisms such as certain virus and bacteria from the highly complex wastewater samples. Secondly, total nucleic acids (RNA/DNA) are purified using the Maxwell® HT Environmental TNA reagents (Promega). Thirdly, RT-qPCR is used to quantify the amount of SARS-CoV-2 in the samples. As the amount of human-derived including faecal matter varies in wastewater depending on, for example, rain, several so-called faecal biomarkers are measured to allow for normalisation. The primary factor for normalisation of SARS-CoV-2 in wastewater is currently the pepper mild mottle virus (PMMoV), which is an abundant virus originating from consumed *Capsicum* spp. The RT-qPCR assays are set up with determined limits of detection (LoD) and of quantification (LoQ) for each target (exemplified in section 5 for SARS-CoV-2). The laboratory workflow is

* Until 31.03.2023, the laboratory analysis was performed by the sub-contracted commercial laboratory Eurofins Environment Testing Denmark, Eurofins Miljø A/S. This change in assay procedure is recorded in the change log accompanying the data.

conducted on a Hamilton VANTAGE (Hamilton Robotics) liquid handling platform, and a more detailed outline of the current full laboratory procedure can be found in section 6.

3: How is the normalised concentration of SARS-CoV-2 in wastewater calculated?

The RT-qPCR laboratory results are expressed as numbers of SARS-CoV-2 and biomarker nucleic acid copies per reaction well; for SARS-CoV-2 and the biomarker (PMMoV) it is RNA copies. All samples are measured as technical triplicates. The copy numbers per well are determined based on dilution series of known standard controls, where the virus concentration has been measured by digital PCR. If technical issues prevent availability of reliable results in less than a full triplicate well set, manual inspection and assessment of data quality of the remaining results is made jointly by the epidemiological and laboratory teams.

For valid samples (see section 6), the average number of copies for each RT-qPCR target is used as follows and illustrated in Figure 3:

- With a positive signal in at least two of the three triplicate wells and an average number of copies ≥3, the average is directly used in the subsequent calculations.
- With a positive signal in at least one of the three triplicate wells and an average number of copies <3, the average number of copies is defined to be 1.5.
- With no signal in any of the triplicate wells, the average number of copies is determined to be 0.25†.

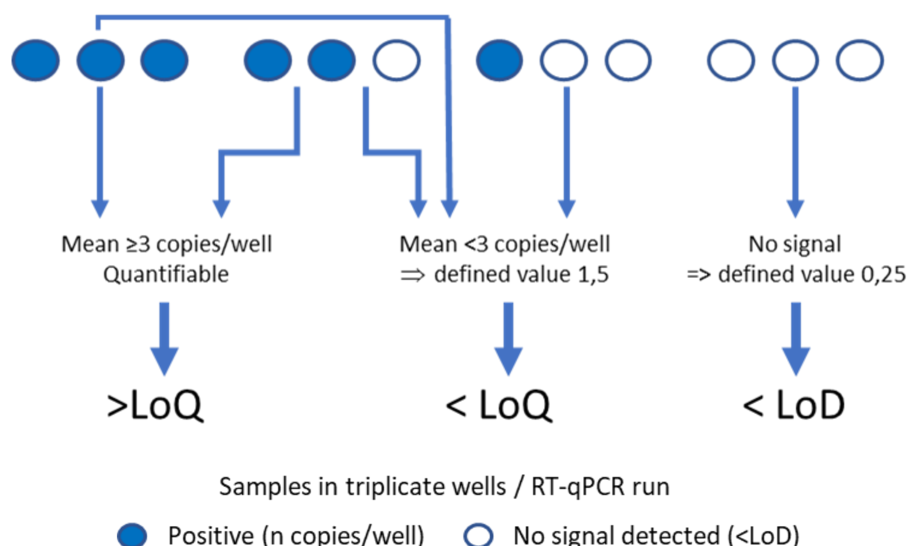

***Figure 3**: Simplified graphical overview over the SARS-CoV-2 nucleocapsid gene detection in the in-house RT-qPCR assay. See Table 6 in section 6 for an overview that includes approval of sample triplicates.*

† As the values in the subsequent calculations are logarithm-transformed a positive number is required, therefore the average number of copies in case of no signal in the triplicate wells is set to 0.25 instead of 0.

The average number of RNA copies per triplicate for each sample is then converted to the number of copies per litre of wastewater using the expression

$$RNA_{copies/litre} = RNA_{copies/\ sample} * 20 * 100$$

where the factor 20 corrects for use of 5 µl per RT-qPCR reaction from a total of 100 µl extract per sample from a start volume of 10 ml. The factor 100 scales up from the initial 10 ml volume to a measure per litre wastewater.

The next step is $\log_{10}$ transformation of the number of RNA copies per sample per litre of wastewater for each target. The $\log_{10}$ transformation is applied to approximate a normal data distribution and to prevent single high measurements (outliers) from disproportionately influencing results, when they are aggregated downstream in the data processing process. For SARS-CoV-2, two regions within the virus' Nucleocapsid (N) gene are measured (N1 and N2) and the average number of gene copies measured per sample is calculated for the $\log_{10}$ transformed copy numbers. To express the amount of SARS-CoV-2 copies relative to the amount of PMMoV (proxy for the amount of human-derived matter in the wastewater) in each sample, the $\log_{10}$ transformed value for PMMoV is substracted. Hence, the following equation is used:

$$\log_{10}(RNA_{mean\ normalised}) = \frac{\log_{10}(N1) + \log_{10}(N2)}{2} - \log_{10}(PMMoV).$$

4: How are the published wastewater results calculated and presented?

The wastewater results are presented for the country (n = 29) and for each of the five Danish regions (n = 4 to 7). See Figure 2 for visualisation of the catchment areas for each region. All calculations related to the wastewater measurements are made on the basis of logarithm-transformed values ($\log_{10}$) and outlier identification is performed based on defined z-score criteria. Two general categories of the wastewater results are used for the weekly published data; weekly weighted averages and trends (growth rates).

4.1 Weekly weighted average

The virus concentration of SARS-CoV-2 in the wastewater per sample is calculated as the number of SARS-CoV-2 RNA copies in relation to the aforementioned biomarker PMMoV (see sections 2 and 3). For data visualisation both at national and regional levels, the results are aggregated by a population weighted average of the viral load results from each wastewater treatment plan. The weighting is done for results from each wastewater treatment plant according to the number of inhabitants in the corresponding catchment area ($\log_{10}$ inhabitants), following the formula:

$$\log_{10}(RNA_{weighted\ average}) = \sum_{i=1}^{n} \log_{10}\left(\mathrm{RNA}_{\mathrm{mean\ normalised}_i}\right)\left(\frac{\log_{10}\left(\mathrm{pop}_i\right)}{\sum_{j=1}^{n}\log_{10}\left(\mathrm{pop}_j\right)}\right)$$

where $\mathrm{RNA}_{\mathrm{mean\ normalised_i}}$ is the concentration measured at site i, pop_i is the number of inhabitants for wastewater treatment plant i and pop_j is the number of inhabitants for wastewater treatment plant j. Before the normalised and weighted average values for SARS-CoV-2 are presented in graphs, the values are back-transformed from $\log_{10}$ scale to normal scale. A scaling factor of 10^7 is used to allow for a reasonable range of values on the y-axis on data visualisations. This gives that

$$\widehat{RNA}_{weighted\ average} = 10^{\log_{10}(RNA_{weighted\ average})} * 10^7 = RNA_{weighted\ average} * 10^7$$

Figure 4 shows an example of population-weighted and normalised SARS-CoV-2 concentrations in wastewater at regional level. Here the light grey shading (95% CI) around the average level (full line) indicates the variable degree of uncertainty of measurements. Variation is derived not only in differences in viral load levels between wastewater treatment plants in a given region, but also from the decreased precision in measurements under either LoQ or LoD (see section 5); as indicated by the broader confidence intervals in the lower section of the graphs.

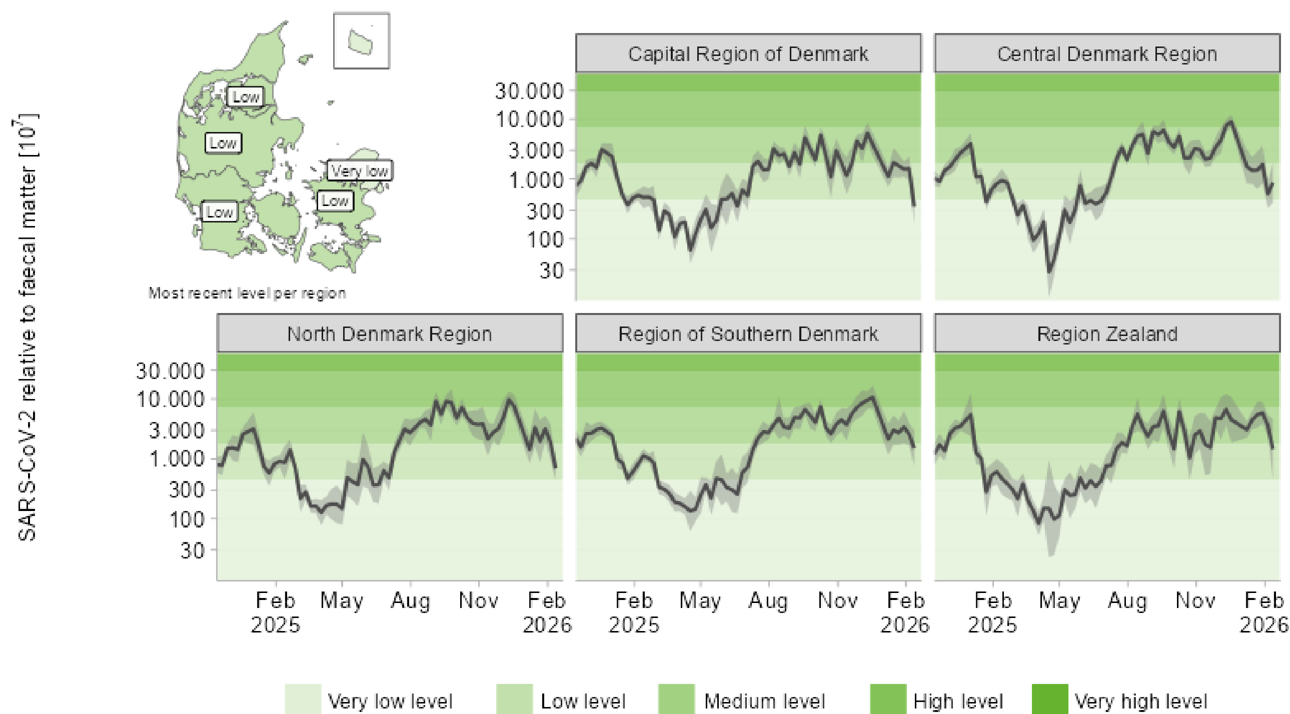

Figure 4: *Example of data aggregation shown for the five Danish regions.*

4.2 Trends (growth rate)

At the national level, a growth rate is calculated to describe the trend in the SARS-CoV-2 concentration based on the last four weeks of data. The growth rate is thus the average weekly change in concentration over the past four weeks. The growth rate is calculated by using a mixed-effects model with wastewater treatment plant as a random effect.

The estimate from the model is converted to an average weekly percentage change as

$$average\ weekly\ growth = (10^{growth\ rate\ *\ 7} - 1) * 100\%$$

The growth rate is classified as one of the seven categories below.

Table 1: Growth rate trend categories.

Very strong increase	≥50%	
Strong increase	25% to 49%	
Increase	10% to 24%	
No growth	-9% to 9%	
Decrease	-24% to -10%	
Strong decrease	-49% to -25%	
Very strong decrease	≤-50%	

The growth rate is shown on visualisations of the normalised concentrations of SARS-CoV-2 in the wastewater at national level (see Figure 5). The growth rates are coloured according to the current trend, and is shown along with the weekly percentage change (see Table 1 above).

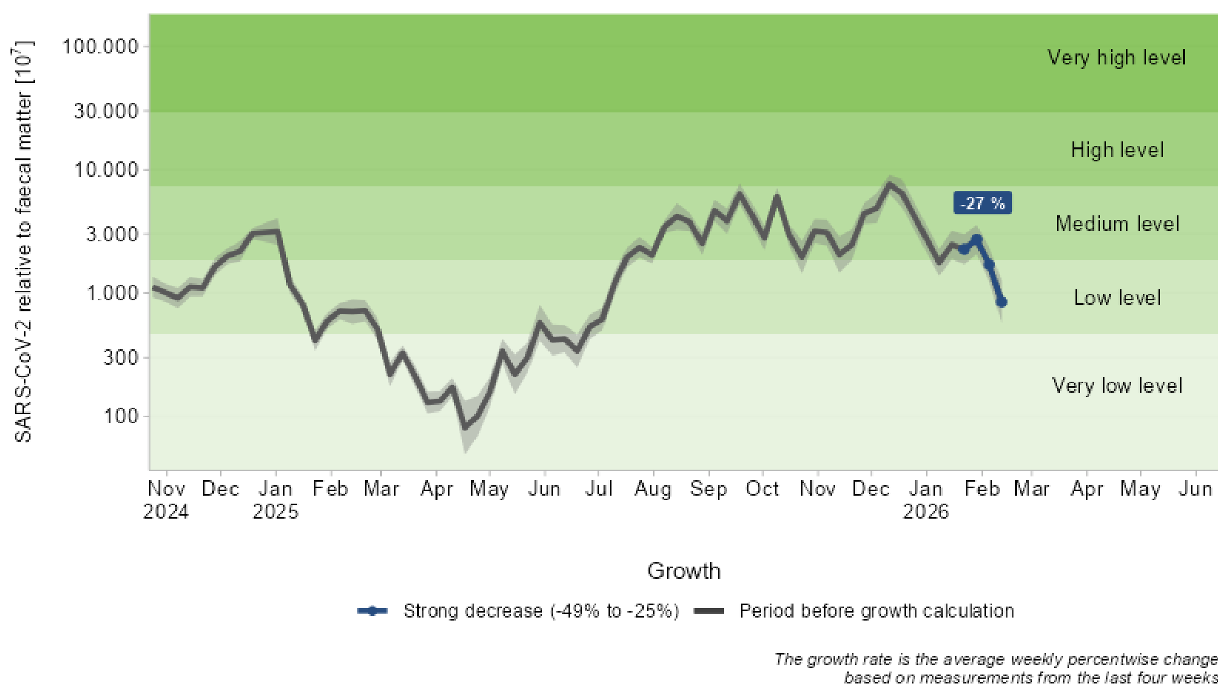

Figure 5: Example of visualisation of population-weighted, averaged and normalised SARS-CoV-2 concentrations (95% CI) in wastewater at national level with growth rate and indicator levels shown. Note that prior to Feb. 1st 2026 sampling took place bi-weekly; the current sampling takes place weekly as described in this document.

4.3 Level assessment

Based on the highest and lowest observed values of the weighted weekly average of SARS-CoV-2 concentrations in the period from 29.06.23 to 07.12.23, five levels were established. Definition of indicator levels were performed based on this period, because it represents a range of suitable high and low concentrations. The boundaries between the levels are evenly distributed on a logarithmic scale and defined so that the lowest and highest concentrations from the period fall into the categories 'Very low level' and 'Very high level', respectively. On the national and regional level graphs, that are updated weekly, the levels are indicated as horizontal bands (Figure 4 and Figure 5).

5: Determination of detection and quantification limits

5.1 What is the limit of detection (LoD) and limit of quantification (LoQ)?

The detection limit is the threshold of gene copies per well above which the RT-qPCR method can reliably detect a signal. The requirement for the detection limit is that detection is expected in at least 95% of the wells.

Meanwhile, the quantification limit is the threshold above which the concentration can be quantified to a defined precision. The requirement for the quantification limit is that the coefficient of variation (CoV) should be below 35%. The CoV is calculated as the standard deviation divided by the mean.
The detection and quantification limits establish the lower bounds for reliable detection and precise quantification of the target(s). For samples, where the average concentration is under the detection or quantification limit, predefined fixed values are used instead of the measured concentrations (as described in section 3).

5.2 Limits for single wells are determined experimentally

In order to determine the detection and quantification limits of the RT-qPCR assay, three independent dilution series were made for each SARS-CoV-2 target, with expected concentrations of 128, 64, 32, 16, 8, 4, 2 and 1 copies per well. For each dilution, 24 technical replicates were made, giving a total of 72 data points per concentration for each gene.
The detection limit was determined as the lowest concentration that met the requirement of detection in 95% of the wells, while the quantification limit was determined as the lowest concentration that met the requirement of CoV below 35% (see Table 2).

Table 2: Experimentally determined detection and quantification limits for the two SARS-CoV-2 genes in single wells.

	Detection limit (copies per well)	Quantification limit (copies per well)
N1 gene	4	8
N2 gene	2	16

As mentioned above, the requirement for the detection limit is detection in 95% of wells. The number of copies in a well follows the Poisson probability distribution. If we impose the requirement that the probability of detection is 95% and apply the Poisson distribution, we find that 3 copies per well will give a signal in 95% of the analysed samples. Thus, 3 is the theoretical expectation for the limit for detection. This aligns well with the results of 4 and 2 copies per well for, respectively, N1 and N2. Therefore, it was decided to use 3 as the detection limit for single wells for both genes.

5.3 Statistical resampling to determine detection limit for triplicate wells

A statistical resampling approach was used to determine the detection limit for triplicates. From the lab dilution series data, wells within each concentration were randomly selected in sets of three to mimic technical triplicates. This was repeated many times, and each set of three was classified as detected or not detected, where detection was defined as a signal in at least one well. The detection limit was then determined as the lowest concentration at which detection occurred in at least 95% of the sets. Based on this analysis, the detection limit for triplicates was determined as an average of 1 copy per well.

5.4 Simulation to determine quantification limit for triplicate wells

As defined above, the quantification limit is chosen so that the CoV is below 35%. According to the Poisson distribution, if the CoV is below 35%, the theoretical expectation for the limit of quantification is approximately 8 copies per well. However, when using the average of triplicates, the variation is reduced. A simulation experiment was made to investigate this reduction in variation and to help determine the quantification limit for the average of triplicates. For a number of concentrations, expressed as the expected number of copies per well, 100 simulations of the calculation of the CoV were made using single measurements and triplicates, respectively. In each simulation, the CoV was determined as the standard deviation divided by the mean of 30 determinations of the number of copies per well.
Figure 6 shows boxplots for the 100 simulations for each concentration and number of replicates. In the dilution experiment with single wells, the quantification limit was determined to be 8 and 16 for the two genes (section 5.2). No tests were carried out with intermediate concentrations. Figure 6 shows that the median at 8 copies per well is close to a CoV below 35%. If the dilution experiment were repeated, quantification limit would be expected to be determined as 8 in half of the cases and 16 otherwise, since intermediate concentrations were not investigated. The experiment is therefore in full accordance with the theoretical expectations. If the calculations of the CoV are instead based on the average of triplicates (grey boxes), the quantification limit becomes 3, as the median here is below 35%.
Based on this simulation, it was decided to use 3 copies per well as the quantification limit when analysing triplicates.

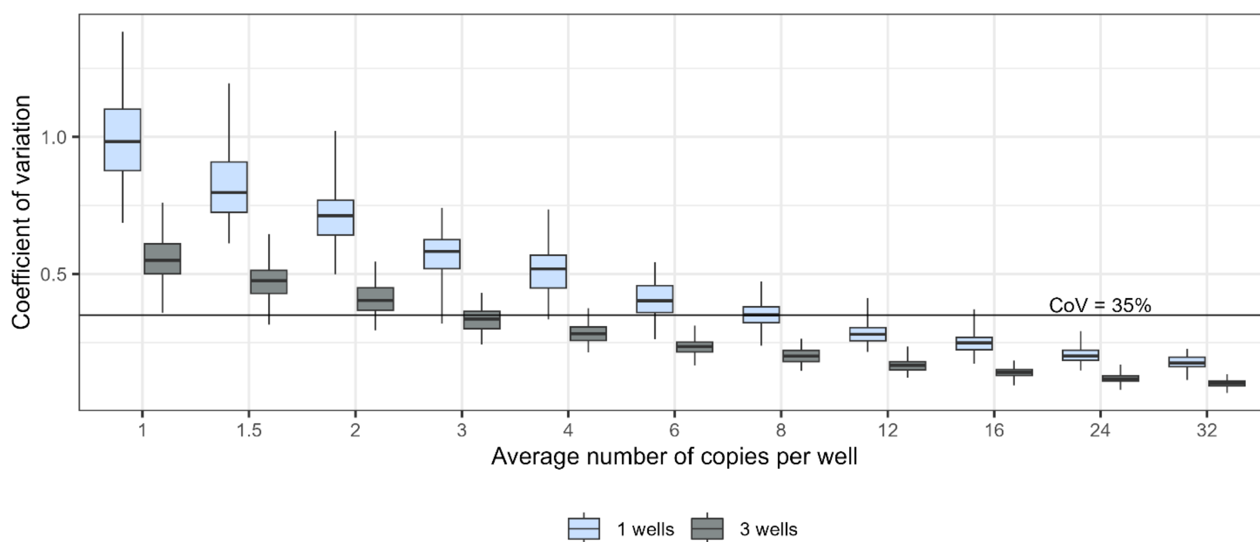

Figure 6: The effect of copy numbers per well, shown for simulations based on single wells (blue) and triplicate wells (grey), on the coefficient of variation (CoV). Each boxplot with interquartile ranges represents 100 simulations per concentration. CoV = 35% is defined as cut-off.

5.5 LoD and LoQ determination for the PMMoV biomarker

As for SARS-CoV-2, the detection and quantification limits of the PMMoV biomarker RT-qPCR assay were determined experimentally in a multiplex setup including three other targets not described here. For

PMMoV the gene encoding the replicon-associated protein is used as target. Standard material was quantified by digital PCR (dPCR) and diluted in 2-fold steps from 500 copies to 0.5 copies per well yielding 11 different concentrations. A total of 3 independent dilution series were prepared. For each dilution series, respectively, 8 and two times 16 technical replicates were analysed resulting in a total 40 data points per concentration. LoD was determined as the lowest concentration, that met the requirement of detection in 95% of the wells. LoQ was determined by calculating concentrations for each dilution using the quantification standard included in each RT-qPCR run at single well level. The coefficient of variation (CoV) was calculated by comparing the RT-qPCR concentration to the expected concentration as determined by dPCR. Simulations based on the experimental data resulted in a quantification limit of an average of 3,4 copies per well based on the simulated sets of triplicate wells.

Table 3: Experimentally determined detection and quantification limits for PMMoV in single wells

	Detection limit (copies per well)	Quantification limit (copies per well)
PMMoV	2	9

6: Laboratory analysis: RNA purification and RT-qPCR

Since 01.04.2023, the laboratory analysis has been performed at SSI‡. All major changes in laboratory procedures are recorded in the change log, that accompany the data available for download.
As outlined in section 2, viral particles are concentrated using NanoTrap® Microbiome A particles (Ceres Nanosciences) from 10 ml wastewater per sample and subsequently total nucleic acids are purified using Maxwell® HT Environmental TNA reagents (Promega). The workflow for RNA concentration, extraction and purification proceeds as follows:

1. Samples are transported and stored at 4°C until analysis starts within 24 hours of collection
2. Pre-aliqouted standardized positive controls are moved from -80°C to the 4°C cold room and handled alongside the samples upon their arrival
3. Samples are mixed manually by inversion and left to sediment for 20 min at room temperature from inversion of the last sample
4. 2x 5mL raw wastewater per sample and positive control are transferred manually to 24-well Whatman Uniplate plates. A negative control is included too.

‡ Until 31.03.2023, the laboratory analysis was performed by the commercial sub-contracted laboratory Eurofins Environment Testing Denmark, Eurofins Miljø A/S.

5. The 24-well plates are transferred to a Hamilton VANTAGE (Hamilton Robotics) liquid handling robot

   All subsequent steps are performed by the liquid handling robot to minimize pipetting errors and reduce repetitive movement manual labor.

6. 50µl Nanotrap® Enhancement Reagent 2 (ER2) is added to each Whatman Uniplate plate well
7. Each sample well is pipette-mixed 10 times
8. 75µl Nanotrap® Microbiome A Beads are added to each well (pre-mix beads just prior to pipetting)
9. The samples are pipette mixed and then incubated for 5 min at room temperature
10. The 24-well Whatman Uniplate plates are transferred to an Alpaqua MagPlate 24 in the robot
11. The sample is left for 11 min for the magnetic Microbiome A beads to settle on the lower well sides in a circle shape
12. The supernatant is carefully removed with a suitable pipette without disturbing the circular pellet
13. The remaining supernatant is removed from the cleared center of the pellet with a smaller pipette
14. The 24-well Whatman Uniplate plates are moved away from the magnet
15. 250uL Cell Lysis Buffer is added to each well
16. Microbiome A beads are resuspended by pipette mixing
17. The 24-well Whatman Uniplate plate is transferred to the Alpaqua MagPlate 24 magnet
18. The sample is left to strip and pellet the beads for three minutes
19. The two supernatants from each sample and control are transferred and combined in a single well in a 96-well Nunc plate
20. 52uL Alkaline protease is added to each well
21. The plate is incubated at 56°C for 20 min with shaking at 1200 RPM
22. 435 uL Resin/Isopropanol mix is added to each well (pre-mix resin thoroughly)
23. The plate is incubated at 56°C for 20 min with shaking at 1200 RPM
24. The 96-Well Deep Well Plate is transferred to a Magnabot® 96 Flex Separation Device
25. The plate is left for 5 min for resin to form a pellet
26. Without disturbing the pellet, the supernatant is carefully discarded
27. The plate is removed from Magnabot® 96 Flex Separation Device
28. 200uL Wash solution is added to each well and the plate shaken on maximum capacity
29. An additional 650uL is added and moderate shaking applied
30. The plate is transferred to the Magnabot® 96 Flex Separation Device
31. The plate is left for 5 min for resin to form a pellet
32. Repeat step 26-31 twice for additional washes
33. Without disturbing the pellet, the supernatant is carefully discarded
34. 450uL 50% EtOH is added to each well
35. Pellet is resuspended by shaking for 30 sec at 1200RPM
36. The 96-Well Deep Well Plate is placed on the Magnabot® 96 Flex Separation Device

37. The plate is left for 5 min for resin to form a pellet
38. Without disturbing the pellet, the supernatant is carefully discarded
39. Let residual Ethanol evaporate by incubation at room temperature for 5min
40. 100uL 25mM Tris-HCl pH8 is added to each well to elute the nucleic acids from the resin
41. Shake the plate for at 40°C for 12min at 1200RPM
42. The 96-Well Deep Well Plate is placed on the Magnabot® 96 Flex Separation Device
43. The plate is left for 5 min for resin to form a pellet
44. The supernatant containing the purified nucleic acids is transferred to a 96-well PCR plate

Extracted nucleic acids are either immediately analyzed by RT-qPCR (as below), or stored at -80°C until subsequent analysis.

10mL aliquoted extraction controls stored at -80°C until use in analysis:
2 positives = pooled wastewater spiked with two different concentrations of cultured SARS-CoV-2 virus.
1 negative = RNAase/DNAase free water (newly dispensed).

6.1 RT-qPCR reactions

Preparation of the RT-qPCR reactions in a 96-well PCR plate is performed on the liquid handling robot by dispensing reagents (mastermix) to relevant wells before transfer of nucleic acid template into the plate. The 96-well plate is then transferred to a Bio-Rad CFX Opus 96 Real-Time PCR System for the RT-qPCR process itself. Data are analysed using Bio-Rad CFX Maestro software.

The concentration of SARS-CoV-2 RNA in the wastewater extracts are determined by RT-qPCR targeting the N1 and N2 regions of the Nucleocapsid (N) gene using the following sequences:

Table 4: *Primer and probe sequences for the N1 and N2 gene regions of SARS-CoV-2.*

Target	Forward primer	Reverse primer	Probe
2019-nCoV_N1	GACCCCAAAATCAGCGAAAT	TCTGGTTACTGCCAGTTGAATCTG	TEX615-ACCCCGCATTACGTTTGGTGGACC-IB-RQ
2019-nCoV_N2	TTACAAACATTGGCCGCAAA	GCGCGACATTCCGAAGAA	FAM-ACAATTTGCCCCCAGCGCTTCAG-IB-FQ

RT-qPCR is performed in 1x Luna® Universal Probe One-Step Reaction mix (NEB) supplemented with 1x Luna WarmStart® RT Enzyme (NEB) and 500µg/µL UltraPure BSA (ThermoFisher) and 5µL template per reaction. The following PCR conditions are used in a total reaction volume of 20µL:

1x	10min at 55°C
1x	3min at 95°C
45x	15sec at 95°C, 30 sec at 60°C

In order to be able to normalise SARS-CoV-2 data to the variable amount of human-derived matter in wastewater, three types of biomarkers are measured. SARS-CoV-2 copy numbers are currently expressed as an amount relative to the PMMoV viral load.

Biomarkers measured in a separate multiplex reaction on 1:100 diluted template:

- Human mitochondrial DNA = MT-CYB
- Pepper Mild Mottle Virus = PMMoV
- CrAssphages = CPQ_056 & CPPol

Table 5: *Primer and probe sequences for the biomarkers measured in wastewater.*

Target	Forward primer	Reverse primer	Probe
MT-CYB	CAATGAATCTGAGGAGGCTAC	CGTGCAAGAATAGGAGGTG	FAM-ACCCTCACACGATTCTTTACCTTTCACT-IBFQ
PMMoV	GAGTGGTTTGACCTTAACGTTTGA	TTGTCGGTTGCAATGCAAGT	HEX-CCTACCGAAGCAAATG-MGB-EDQ
CPQ_056	CAGAAGTACAAACTCCTAAAAAACGTAGAG	GATGACCAATAAACAAGCCATTAGC	TEX615-AAT+AACGA+TTTA+CGT+GAT+GTAA+CTC-IBRQ
CPPol	ATGTWGGTARACAATTTCATGTAGAAG	TCATCAAGACTATTAATAACDGTNACAACA	Cy5-ACCAGCMGCCATTCTACTACGAGHAC-IBRQ

RT-qPCR is performed in 1x Luna® Universal Probe One-Step Reaction mix (NEB) supplemented with 1x Luna WarmStart® RT Enzyme (NEB) and 500µg/µL UltraPure BSA (ThermoFisher) and 5µL (1:100) template per reaction. The following PCR conditions are used in a total reaction volume of 20µL:

1x 10min at 55°C
1x 3min at 95°C
45x 15sec at 95°C, 30 sec at 58°C

On each RT-qPCR plate, a five-fold 10x dilution of a quantification standard from 200.000 copies / PCR reaction to 20 copies / PCR reaction for each target is included in technical triplicates. The concentration of the quantification standard is independently verified using digital PCR. Additionally, a non-template control and a positive control of double stranded synthetic DNA (Gblock) carrying the target sequences of relevance and of known concentration is also included in each RT-qPCR run. All wastewater samples as well as positive and negative extraction controls are measured in technical triplicates.
For each sample the results per triplicate are evaluated. If samples do not fulfil the criteria (Table 6), they are assessed with potential technical errors in mind and decisions as to whether to rerun a sample or exclude the result(s) are made through dialogue between the laboratory and epidemiological teams.

***Table 6**: Overview over possible combinations of target copies per well for non-approved samples and the associated result. Situations that result in valid samples are shown in Figure 3. Subsampling is from the same extraction for each triplicate (5 ul pipetted/well).*

Copies/well 1	Copies/well 2	Copies/well 3	Outcome
0	0	≥ 6	Flag to lab; highly unlikely Rerun or exclude
0	pos	pos	Average of the positive wells ≥ 6 Flag to lab; highly unlikely. Rerun or exclude
0	0	X	Flag to lab; Rerun or exclude
0	≥ 6	X	Flag to lab; highly unlikely Rerun or exclude
0	X	X	Flag to lab; Rerun or exclude
≥1	X	X	Flag to lab; Rerun or exclude

0 = no signal.

X = technical issue, no read-out from well.

7: What data are shared?

You can download the latest data on SARS-CoV-2 detection in wastewater on our website. The data is collated in a zipped folder containing the following files:

Format	File name	Content
.pdf	Readme-wastewater	Overview over variables in the datasets
.csv	20XX-xx-xx_dk_wastewater_data	National level data
.csv	20XX-xx-xx_region_wastewater_data	Regional level data
.html	20XX-xx-xx_wastewater_graphs	Visualisation of aggregated data
.txt	changelog-wastewater	Overview of all procedural changes that have relevance for the delivered data and analyses
.txt	Operationsstatuslog-wastewater	Overview of all major operational changes that may have relevance for data analyses

For further information, the SSI wastewater team can be contacted on spildevand@ssi.dk.