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Summary of preliminary results from wastewater analysis for tracing SARS-CoV-2 in Stockholm region

Background and method

Sampling of wastewater has been done since mid-April at Bromma, Henriksdal, and Käppala wastewater treatment plants (WWTP). These treatment plants receive wastewater from a population of approximately 360,000; 860,000 and 500,000, respectively.

The sampling at Henriksdal and Bromma started in week no. 16 (13 April) and the Käppala sampling started in week no. 18, following a sampling protocol developed by KTH. The flow-proportional pooled samples were taken bi-weekly until end of July. From week no.35 the sampling was done every week. After concentration, filtering and preparation, the samples have been analyzed using qPCR technique for genetic material (RNA) belonging to the virus SARS-CoV-2, known to cause the COVID-19 pandemic.

When analyses have not been possible immediately, raw wastewater has been frozen at –20 degrees, and concentrated wastewater or purified RNA stored at -80 C, until the next analysis step has been carried out.

During June and July, KTH researchers have compared four different concentration methods, two of them are commonly used internationally and the other two were modified methods adapted by the KTH team. The sensitivity of two ultrafiltration-based methods and two adsorption and extraction-based methods were compared for the SARS-CoV-2 as well as for two reference viruses. Our investigation concluded that the double ultrafiltration method adapted by KTH has a significantly higher efficiency compared to single filtration and adsorption methods. The scientific article has been published in Science of Total Environment, an internationally leading scientific journal, and is available on this link.

We wish to re-iterate that water and wastewater are not known to be important pathways for transmission of the virus. We have not detected any virus in treated outgoing water and the tracing of SARS-CoV-2 in wastewater should not cause any need for additional precautions for water consumers.

Preliminary Results from SARS-CoV-2 monitoring in Stockholm Region

The RNA signal detected in the wastewater analyses reflects the amount of virus excreted from infected persons within the catchment area, and may thus be used as an indicator when assessing the trend of the COVID-19 pandemic among the population. The analysis results have been converted into gene copy numbers.

- From end of April to middle of May, coinciding with the end of the first COVID-19 wave in Stockholm, we detected the genome of SARS-CoV-2 in the wastewater.

- From July 13 to August 23 we were not able to detect the RNA from SARS-CoV-2 in any of the samples. At this time, Stockholm had a low spread and only single ICU cases.

- At the end of August, the known cases had not increased, but the virus was again detected in the wastewater. During mid-September peak levels were detected, especially from the Käppala wastewater plant (week 38). This was right before clinical cases started to increase. We interpret the increasing levels in August and early September as an early warning of coming spread, but this needs further validation.

- During week 41-43, the viral genome was detected in some but not all inlets. These results deviated from the expected trend as the numbers of reported cases and intensive care unit cases in the region now were increasing steadily.

- The decrease in the detected virus coincides with a period when analyses had to be postponed due to lack of laboratory supplies. Samples were frozen and then analyzed at a later point when materials supplies had resumed. One hypothesis is that the storing of samples by freezing negatively affects the possibility to detect the virus, which is also supported by recent international experience. This requires further investigations.

- From week 44, continuous analyses have been performed immediately on incoming water, without storage and freezing of samples. These analyses have been done in triplicates, and yielded consistent results. Other improvements include the normalization of test results against an internal (and harmless) virus commonly found in wastewater, thus reducing effect from stormwater dilution and variations in the wastewater composition.

- During week 44 to 48 the detected genomic level has again risen, and remain at peak level. The level has been particular high from the Henriksdal wastewater inlet uptake area.

There are fluctuations in the observed genomic level which remains to be scientifically explained, before certain comparisons over time can be made with a high degree of certainty. With the improvements made in the sample management and analyses we believe our results are more accurate and significant.

While this is a research project, and not yet validated as a tool for decision making, monitoring the virus in the wastewater can still give useful information about the spread of the virus among the population in Stockholm region. This can complement other sources of information. As the pandemic appears to persist well into 2021, the monitoring of virus prevalence in the 1.7 million people within the three wastewater treatment plants may provide important additional information to the knowledge basis for decision-making, such as future decisions on possible lifting of restrictions.

During the month of October 2020 the Region of Stockholm decided to support our research with 1 MSEK in order to continue the sampling and to improve wastewater-based epidemiology methods and tools. KTH is grateful for this support and for our collaboration with the Region.

We also welcome the continued collaboration with Stockholm Vatten och Avfall AB, Käppalaverket, SciLifeLab, Uppsala University and SLU, and international partners, in developing knowledge and methods for wastewater-based epidemiology.

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