COULD HIGH-THROUGHPUT WASTEWATER ANALYSIS BECOME A KEY TOOL IN SARS-COV-2 SURVEILLANCE?

Introduction

As governments and public health organisations globally use prevention and control methods to stop the spread of COVID-19, strategies to monitor the pandemic have been a primary area of focus. In many countries, the COVID-19 pandemic has been exacerbated by the implementation of unoptimised testing strategies. Antigen and PCR testing, for example, has largely proven insufficient to monitor disease transmission in local communities, primarily because testing strategies often do not include systematic testing of the general population, making it challenging to monitor asymptomatic patients. As such, complementary methods have been implemented by many authorities around the world to provide more holistic information about public health.

Monitoring wastewater has been successfully used as one such method to detect SARS-CoV-2 levels in local communities [1]. SARS-CoV-2 can be shed in the faeces of individuals with symptomatic or asymptomatic infection, meaning wastewater surveillance is able to capture data on both types of infection. The fluctuation of viral levels in sewage systems provides an indirect early warning system of COVID-19 outbreaks in the surrounding communities, and can assess how preventative methods of vaccination drives and social distancing are reducing transmission.

While this method does not provide detailed information about how many people have the virus or who they are, it does give population-level oversight of SARS-CoV-2 levels with the benefit of a readily accessible testing sample. Sewage water samples can then be tested in local COVID-19 laboratories to inform public health decision-making.

Wastewater-based epidemiology (WBE) programmes traditionally use wet chemistry methods to measure several biomarkers of COVID-19 in addition to viral levels of SARS-CoV-2 to give more comprehensive information about public health. Effective SARS-CoV-2 surveillance needs accurate results for large numbers of samples, but WBE workflows are often time-consuming, labour-intensive and costly, meaning the potential for tracking COVID-19 in water samples cannot be utilised by many laboratories. This article discusses how high-throughput and cost-effective discrete analysis workflows are overcoming these limitations, enabling testing laboratories to monitor infection rates, and the spread or control of SARS-CoV-2 in communities.

How can wastewater testing contribute to the control of COVID-19?

SARS-CoV-2 detection in wastewater can indicate the prevalence of COVID-19 in a community, and when used over time, show how infection levels respond to preventative measures. The National Wastewater Surveillance System (NWSS) in the U.S. is already seeing infection extent and trend information, with wastewater viral concentration levels closely matching the number of new cases in an area [2].

Laboratories mainly rely on quantitative reverse transcription polymerase chain reaction (RT-qPCR) techniques to measure and quantify SARS-CoV-2 RNA in wastewater, although several variables can make this challenging. For example, viral load measurement of SARS-CoV-2 is critical, and survival of the virus in water depends on temperature, pH, concentration of suspended solids and organic matter, and levels of disinfectant. As such, complementary methods to provide more comprehensive and contextualised population data are required for a more accurate measurement of infection within a community.

Measurements of several chemical and physiochemical biomarkers provide longitudinal information about community-wide health conditions that can be tracked and compared with COVID-19 viral data. These biomarkers include total nitrogen, phosphorus, ammoniacal nitrogen, urea and creatinine, and indicate normal faecal load and seasonal or daily fluctuations against which viral data can be analysed. Additionally, markers, such as pH, conductivity, ammonia, nitrate, nitrite, total organic nitrogen, ortho-phosphate, and other ions such as chloride and sulphate, can provide indirect information about the survival period of a virus within the wastewater.

Traditional techniques can't keep up with the throughput

Together, RT-qPCR measurement of SARS-CoV-2 and wet chemistry measurement of complementary biomarkers is an effective way of assessing the level of infection within communities. However, these methods are time-consuming and costly, which can severely limit the capability of local authorities to track COVID-19 infection in wastewater.

WBE programmes in testing laboratories rely on complex workflows with several moving parts that must integrate to be successful. For example, specialist scientists are required to run and monitor each test, and each test requires multiple methods with sequential steps for each parameter, all needing separate samples and large reagent volumes for each sample. A typical determination of ammonia by titration not only requires a large sample volume (~100 ml) to reach appropriate limits of detection but also requires glassware and an adequate cleaning process to avoid contamination, creating barriers to rapid testing of high numbers of samples. Using hazardous reagents containing mercury means a mercury recovery procedure must be included for any waste and large volumes of acid may also be required, exposing the analyst to unnecessary and avoidable risks. Greater statistical variance between analysts during the method validation process results in a higher limit of detection and therefore a narrower analytical range for which parameters such as flow and faecal load can be estimated against.

Another challenge is to do with how samples are collected in the field and how this affects bulk testing in wet-chemistry labs. For SARS-CoV-2, seven day monitoring of wastewater samples is essential if trends and patterns in data are to be identified and interpreted correctly whilst the data is still relevant and a local government can respond in time. Often, scientists find that it is only possible to take a quick 'grab sample' than a more representative composite from sampling locations due to the large number of sites needing to be tested. 'Grab samples' are more challenging to analyse as it is important to monitor complimentary analytes such as ammonia, phosphate, pH, and conductivity in order to estimate a correction of the PCR results. The ammonia result is used to adjust for flow and population input, adding context and perspective to the data. Phosphate, pH, conductivity and suspended solids can provide additional and confirmatory information about the sample, as well as acting as qualifiers for the ammonia result. It is therefore important that rapid repeat analysis is performed to confirm suspect or erroneous results – something that simply isn't feasible with traditional wet-chemistry techniques when dealing with hundreds of samples per day.

With slow processes like these, labour-intensive and costly workstreams often cannot deliver the high throughputs needed for continuous or rapid testing to assess infection levels.

Could consolidated, multiparameter, discrete analysis hold the key to effective wastewater surveillance?

Fortunately, next-generation discrete analysers are providing testing laboratories with the solution for high-throughput testing of the entire range of SARS-CoV-2 biomarkers in wastewater samples. With these new technologies, multiple biomarker parameters in water samples can be analysed in one machine quickly and easily using automated methods. These systems were developed specifically to increase throughput of sample analysis with high accuracy, and simplifies the wet chemical analysis to offer flexibility and efficiency.

Collected samples can be tested for chemical and population size markers in compliance with internationally approved standard methods, including DIN, ISO and U.S. Environmental Protection Agency (EPA). Using EPA methods, for example, users can measure ammonia levels in a range of 0.01 to 2.0mg/L NH_3. Coupled to this, some analysers have modules that are able to conduct electrochemical analysis for pH and conductivity in sewage water samples. The benefit here is that these systems can perform automated analysis of multiple parameters in parallel, saving valuable time.

Moreover, automated functionality means one operator can run up to 20 parameter tests on a single sample and walk away while these tests are running. Some machines can analyse more than 100 samples and 40 reagents in one run, with up to 350 photometric tests and up to 67 ECM measurements completed in an hour. With this automation, the risk of manual preparation error is removed, while analysis is simplified and accelerated.

In SARS-CoV-2 surveillance, fast, frequent and continuous monitoring of chemical and physiochemical biomarkers is critical to establishing accurate insight. Discrete analysers deliver fast biomarker identification and quantification with the added benefits of increased accuracy, reduced labour, fewer reagents and decreased costs.

Quick and continuous monitoring will win the fight against COVID-19

As governments and public health agencies across the world fight to control COVID-19, WBE programmes are forming a large part of the battle plan. RT-qPCR tests, in combination with biomarker surveillance, can detect SARS-CoV-2 and estimate circulation, even when infected individuals are asymptomatic. Moreover, when sampling is performed frequently, this method can track COVID-19 rates long-term.

Traditional wet chemical analysis does not meet the fast and continuous requirements for this surveillance due to large labour, time and cost demands. With consolidated, multiparameter testing, next-generation discrete analysers can provide the fast and accurate biomarker detection and quantification needed to understand population infection rates. By simplifying and automating tests, and producing accurate and reproducible biomarker results, significant savings can be made in labour, time and costs. Together, these benefits provide the effective and continuous wastewater surveillance needed to help combat the ongoing COVID-19 pandemic, as well as any potential future pandemics or large-scale infections.

References

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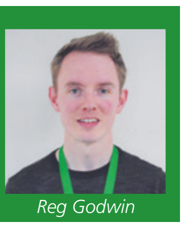

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