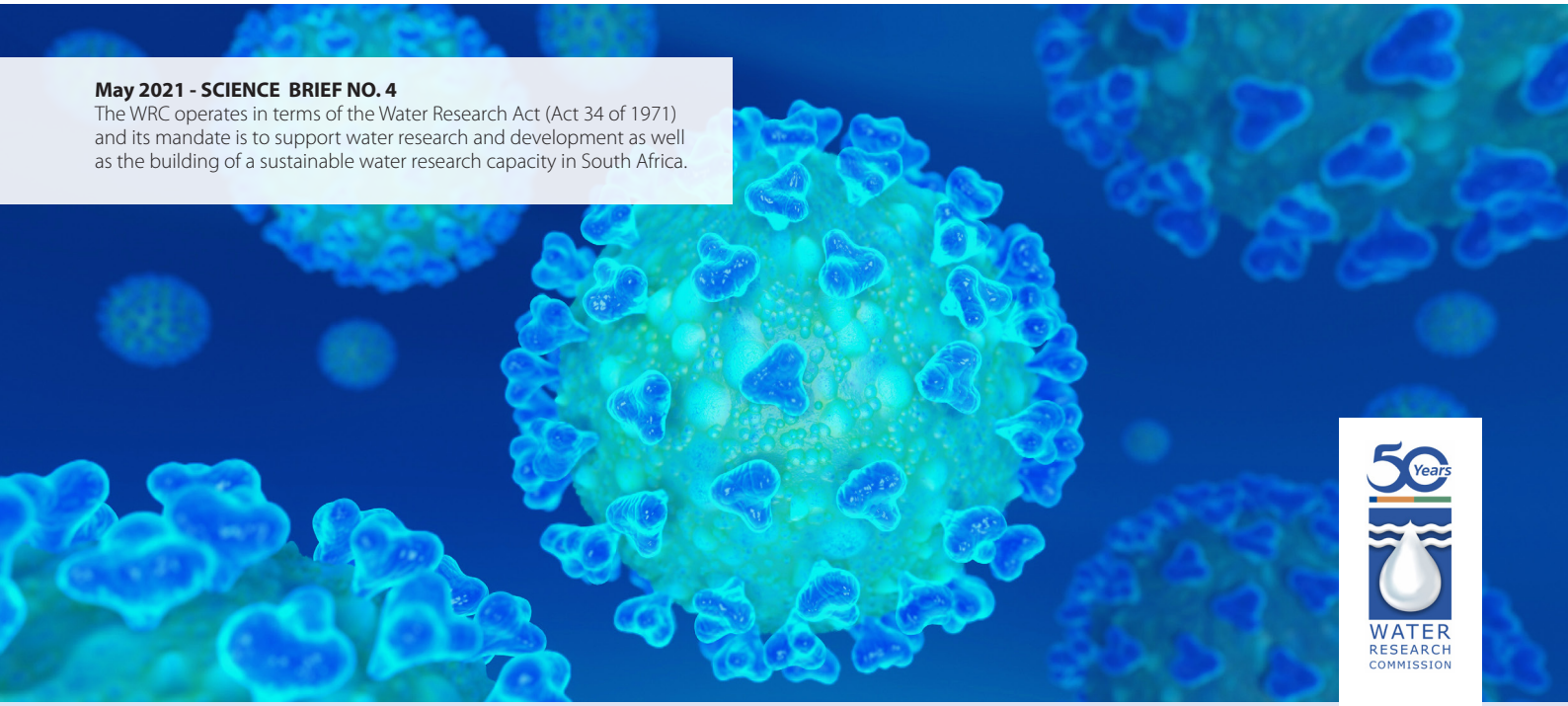

May 2021 - SCIENCE BRIEF NO. 4
The WRC operates in terms of the Water Research Act (Act 34 of 1971) and its mandate is to support water research and development as well as the building of a sustainable water research capacity in South Africa.

Wastewater Surveillance of the SARS- COV-2 virus indicates the start of the third wave of infections

The first important finding from the interpretation of the data and information on testing for COVID-19 RNA fragments in wastewater confirms that wastewater-based epidemiological (WBE) surveillance is a cost-effective means of providing an early warning of the spread and increase in infections. In this case the RNA signal in the wastewater showed increases in copy numbers, at least 3 weeks in advance before the clinical cases were starting to confirm increases in reported positive cases.

The second important finding is that the increase in the viral loads or increase in the amplification of the RNA signal offers a very effective signal on the increases in infections, as it is able to capture the asymptomatic cases as well which form a pathway for the spread of infections.

This progress has demonstrated the importance and effectiveness of WBE surveillance and why it needs to be escalated at a national level in the fight of this pandemic, but also in the future be used to determine the impact of the large rollout of vaccinations in the country.

Introduction

The concept of screening municipal wastewater and environmental water quality as an epidemiological tool for viruses is not a new concept and has been used to help inform broader infectious disease epidemiological surveillance and mitigation efforts such as the Global Polio Eradication Initiative. Environmental surveillance has also been used and recommended for other infections, such as typhoid, early warning of hepatitis A and norovirus outbreaks, as well as for antimicrobial resistance, with modelling techniques used to assist both the design and interpretation of those efforts. Wastewater-based epidemiology (WBE) is also commonly used in the surveillance of licit and illicit drugs and various chemical contaminants which may impact human health[8]. Recent studies have shown that environmental surveillance of SARS-CoV-2 (the virus responsible for the COVID-19 disease) signals or biomarkers (typically RNA) by means of Reverse transcriptase Polymerase Chain Reaction (Rt-PCR) RNA by means of Reverse transcriptase Polymerase Chain Reaction (Rt-PCR) can be a low-cost solution for tracking Covid-19 outbreaks in communities.

This is because SARS-CoV-2 is shed at relatively high titres in the stool of some individuals. Viral gastrointestinal infection, or at least shedding, can remain for some time after clearance of the virus in the respiratory tract. The detection of SARS-CoV-2 RNA in untreated domestic wastewater was

first reported internationally by KWR-Netherlands and this had spurned many other countries into the pathway of WBE (more than 100 countries have some form of formalised activity). The detection of SARS-CoV-2 in sewage is relevant not necessarily because of the potential risk of disease spread, but rather because of the potential to determine the presence of infected individuals in a community. SARS-CoV-2 screening in raw sewage water using Rt-PCR can therefore be used as a tool to measure the virus circulation in a defined population, for example a city or a smaller municipality feeding to the same wastewater treatment works.

National SARS-COV-2 Wastewater Surveillance Initiative

In response, the WRC, in partnership with SALGA, launched the Water Quality (wastewater and non-sewer) National surveillance programme aimed to complement national initiatives in dealing with the pandemic. The intent of this programme is to facilitate the implementation of a nationwide initiative for the surveillance of COVID-19 spread in South African communities using a water and sanitation-focused approach. Through this process contribute to supporting the national response to the pandemic by serving as hotspots identification and an early warning for the resurgence of the virus and other waterborne disease outbreaks.

For this reason, implementation of the wastewater surveillance initiative followed a three-phased approach, with **Phase 1** being the proof of concept aimed at optimising sample design, testing and fine-tuning sampling protocol, preliminary sampling and analysis of wastewater samples from selected metropolitan cities, which was completed in September 2020 and the results afforded the opportunity to launch into phase 2 in late October 2020. P**hase 2 (**pilot-scale monitoring), funded by the WRC, is in progress, and builds partnerships for pilot-scale monitoring which establishes and will see the commissioning of a collaborative monitoring initiative in provincial hotspots (Gauteng, KwaZulu-Natal and Western Cape) using the sampling and testing protocols developed in phase 1. Phase 2 is now led and coordinated by the National Institute of Communicable Diseases (NICD) in partnership with SACCESS (South African Collaboration COVID-19 Environmental Surveillance System). **Phase 3** is the incremental growth towards national wastewater surveillance where most of municipalities and treatments plants get monitored.

In parallel, the WRC has launched and is funding several research activities that contribute to managing this pandemic in the long term, namely:

- Ecosystem responses to the large-scale use of SARS-CoV-2 disinfectants
- Non-sewered surveillance programme: sampling of non-sewered areas and rivers as a non-invasive, preliminary surveillance method to establish the spread of SARS-CoV-2 in South African communities
- Evaluation of health risks associated with occupational exposures to biological and chemical contaminants at wastewater treatment plants and recycled water use sites
- Tracking the evolution of SARS-CoV-2 and the emergence of other infectious diseases in communities using a wastewater-based epidemiology approach
- Genomic variants studies
- Advanced molecular method for the detection and quantification of SARS-CoV-2 in wastewater and sanitation environments

The Durban University of Technology (DUT)– Institute of water and wastewater technology (IWWT) progress

The research being undertaken by DUT-IWWT is supported by the WRC, in partnership with eThekwini Municipality and Umgeni Water. The project is aimed at enhancing the techniques and methods for sampling and testing of the SARS-CoV-2 RNA fragments in wastewater. This has allowed the DUT to monitor the treatment plants in the Durban area since July 2020. The Central Wastewater Treatment Plant serving a large number of suburbs (nearly 65 including the CBD) is used as a case study. The continued monitoring has resulted in significant findings and confirmed some of the early positions of the initiative as presented below:

Detection and quantification of the N2 gene within the SARS-CoV-2 genome has been ongoing at the head of works at Central Wastewater Treatment Plant (CWWTP) since July 2020

The CWWTP services approx. 61 suburbs (Appendix 1) within the eThekwini Municipality and is a good representation of KZN as a whole (Figure 1)

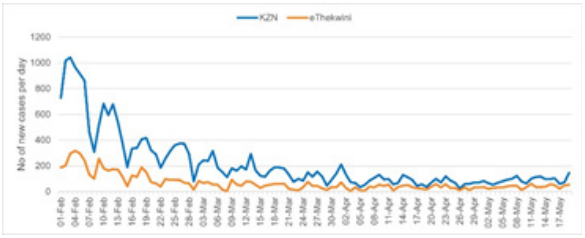

Figure 1: No. of new clinical cases per day in KZN Province

The number of active clinical cases in KZN and eThekwini Municipality has been increasing steadily since 20 April 2021, indicating that another wave of COVID-19 is possibly emerging (Figure 2)

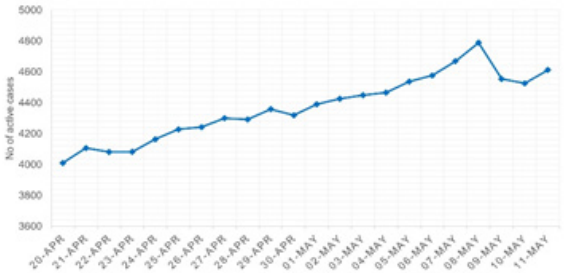

Figure 2: No. of active cases in KZN

Wastewater surveillance revealed an increase in (SARS-CoV-2) copy numbers as early as 30 March 2021 – 3 weeks before reported clinical cases (Figure 3)

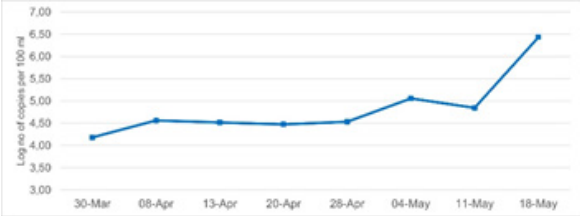

Figure 3: Increase in SARS COV2 viral load in wastewater March to May 2021 (Source: DUT-IWWT)

Previous and current data indicate that the easing of lockdown restrictions (level 3 to level 1) contributes significantly to the increased viral loads in wastewater (Figure 4)

Figure 4: Monitoring of CWTTP Jan – May to May 2021 (Source: DUT-IWWT)

The number of active clinical cases in April/May 2021 is significantly lower when compared to February 2021 despite the viral loads in wastewater being almost the same (Figure 4). This suggests that there may be a greater number of infected individuals within the population than what is being reported currently for eThekwini.

There has been a significant increase in SARS-CoV-2 copy numbers in wastewater (Table 1 and Figure 3) observed over the last 4 weeks (28 April 2021 – 18 May 2021) indicating that a spike in clinical cases in the coming weeks is imminent – and requires urgent attention from the relevant authorities.

Date	Copies per 100 ml of wastewater
30 March	15100
08 April	36300
13 April	33000
20 April	29872
28 April	34112
04 May	117000
11 May	70000
18 May	2710000

Table 1: Quantification of SARS-CoV-2 viral load in wastewater (March – May 2021)

Conclusions

The first important finding from the interpretation of the data and information on testing for COVID-19 RNA fragments in wastewater, confirms that wastewater-based repidemiological (WBE) surveillance is a cost-effective means of providing an early warning of the spread and increase in infections. ***In this case the RNA signal in the wastewater showed increases in copy numbers, at least 3 weeks in advance before the clinical cases were starting to confirm increases in reported positive cases.***

The second important finding is the increase in the viral loads or increase in the amplification of the RNA signal offers a very effective signal on the increases in infections, as it is able to capture the asymptomatic cases as well which form a pathway for the spread of infections.

This progress has demonstrated the importance and effectiveness of WBE surveillance and why it needs to be escalated at a national level in the fight of this pandemic, but also in the future be used to determine the impact of the large rollout of vaccinations in the country.

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COVID-19 is an issue of national security and wastewater-based epidemiology contributes to the aspect of security. Vaccines and other interventions are more immediate, however, WBE plays a longer term role as well in tracking the virus and tracking any future waves.

This report has been prepared by the Durban Institute of Technology – Institute of water and wastewater Technology (DUT-IWWT) Research Team led by Prof Faizal Bux and Jay Bhagwan of the Water Research Commission (WRC).