

National Wastewater Surveillance Programme - COVID-19

Weeks 38 & 39 (Weeks Ending 25 September & 02 October 2022)

Report prepared on 05 October 2022

99%

sites tested in the last 2 weeks had SARS-CoV-2 detected (102/103 sites)

69%

NZ population covered by wastewater testing

Omicron BA.4/5 (>93%)

Most prevalent variant detected

Nationally, there was an increase in SARS-CoV-2 levels in wastewater in week 39 compared to week 38. This does not indicate a sustained increase.

In the most recent week (week 39, ending 02 October 2022), 9% of sites have levels of SARS-CoV-2 that were either below the limit of quantitation or not detected. In week 39 compared to week 38, 49% of sites showed increased SARS-CoV-2 levels and 28% of sites showed a decrease. For week 39 compared to a month ago (week 35, ending 4 September 2022), 49% of sites showed increased levels and 33% of sites showed a decrease. The results for weeks 38 and 39 suggest that reported case rates are 26% and 40% lower than would be expected based on the SARS-CoV-2 levels in wastewater, respectively.

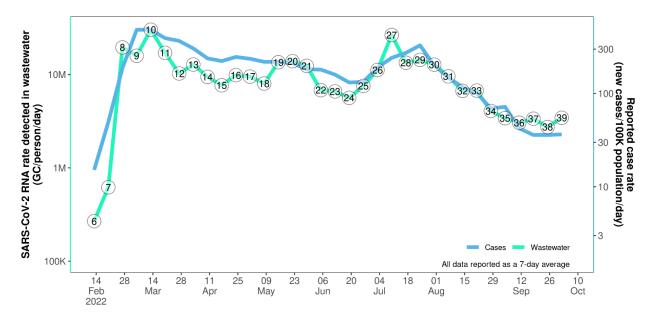


Figure 1. National timeseries of estimated SARS-CoV-2 genome copies (GC) in wastewater rate (GC/person/day, green line) and reported case rate (new cases/100,000 population/day, blue line). Numbers in the points are the week of the year. Log₁₀ scale. Data reported as 7-day average.

Combined Results for Weeks 38 & 39 (Weeks ending 25 September & 2 October 2022)

In the two weeks ending 2 October 2022, 274 samples were collected from 103 locations across New Zealand.

SARS-CoV-2 RNA was **detected** in 270 samples from 102/103 (99%) sites tested during this period (Figure 2, Table 1). SARS-CoV-2 RNA was **not detected** in four samples collected from Gore, Otane, Woodville and Waverley. However, all these sites returned a second positive sample within the same week except for Waverley, for which both samples were negative in week 38.

Please note that due to a calculation error in Figure 2, the proportions of 'detected, not detected and detected (not quantifiable)' for weeks 36 and 37 have now been corrected.

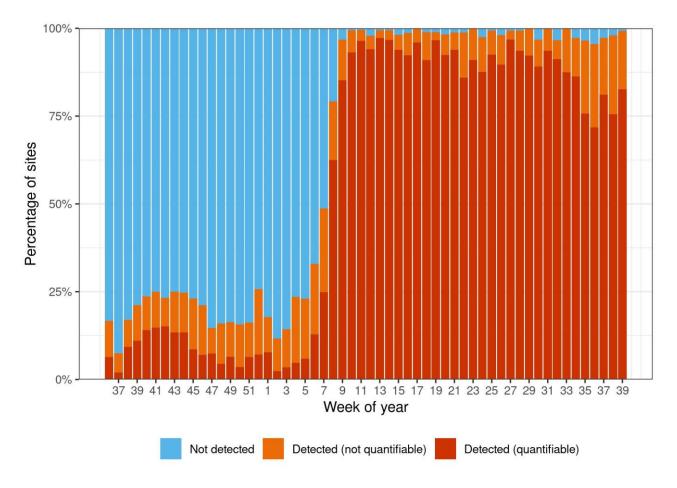


Figure 2. Results for SARS-CoV-2 RNA in wastewater collected across New Zealand, showing the proportion of sites with quantifiable levels of SARS-CoV-2 RNA (red), not quantifiable levels (orange) and no SARS-CoV-2 RNA detected (blue).

Regional Trends

Regional summaries (Figure 3) of the wastewater data indicates steady or increasing viral levels across all regions except Auckland Metro, which shows an ongoing reduction. For those regions where viral levels have increased this week, it is too early to determine whether this is a sustained increase.



Figure 3. Total SARS-CoV-2 genome copies detected per day in the five Ministry of Health regions (Rolling two-week average).

There is however site to site variation as illustrated in Figure 4 and the individual site plots. In week 39 compared to week 38, 49% of sites showed increased SARS-CoV-2 levels and 28% of sites showed decreased levels (Figure 4A). For week 39 compared to a month ago (week 35), 49% of sites had an increase in levels and 33% of sites had a decrease (Figure 4C). Sites with sustained increases (more than two weeks) include Hutt Valley, Wellington (Moa Point), Wellington (Western), Picton, Rolleston, Timaru, Ashburton, Leeston, Invercargill and Bluff.

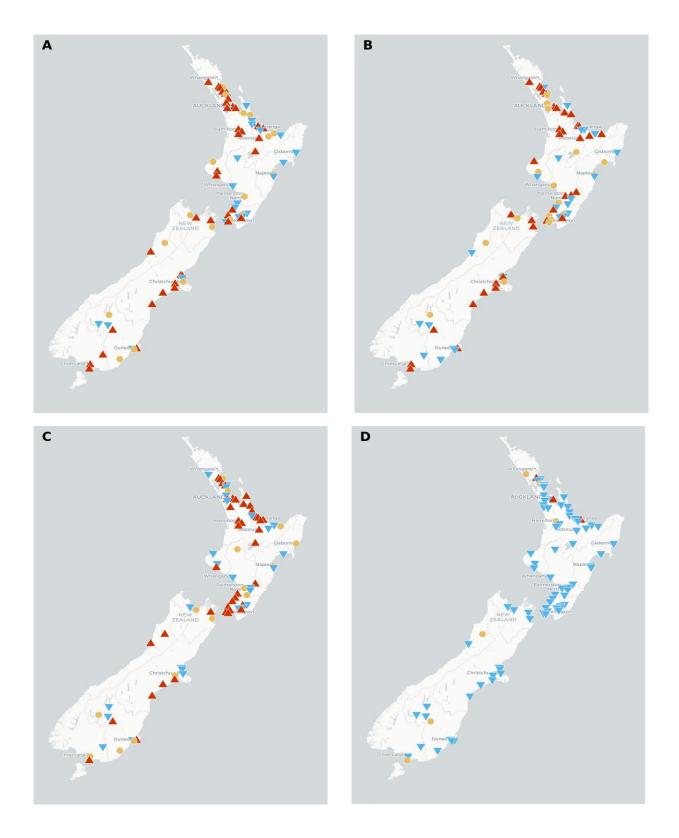


Figure 4. Comparison of SARS-CoV-2 levels for the week ending 2 October 2022, compared to levels measured: A) 1 week ago; B) 2 weeks ago; C) 4 weeks ago; D) 12 weeks ago. Only sites with results for both time points are included. When the viral quantity is 30% or more higher this is labelled as increased (red up arrow on map). When the viral quantity is 30% or more lower, this is labelled as decreased (blue down arrow on map). If viral levels have changed less than this in the compared weeks, this is labelled as no change (yellow circle on map). Interactive map of weekly results available publicly at https://www.esr.cri.nz/our-expertise/covid-19-response/wastewater-testing-results.

Wastewater Variant Analysis

In collaboration with Wilderlab, ESR generated the variant analysis results (Figure 5, Figure 6) from a set of nationwide sentinel sites in: Week 38 (ending 25 September 2022) and Week 39 (ending 2 October 2022).

The overall decrease in case numbers is reflected in decreasing viral loads in wastewater samples. This can make variant detection and interpretation more difficult. The level of precision and sensitivity in these percentage estimates can be uncertain.

The BA.4 and BA.5 variants are the dominant circulating variants across Aotearoa (national average of 93%). In the past fortnight 81% of monitored sites were 100% BA.4/5.

Following possible low-level detection of BA.2.75 in Mt Maunganui in week 34, this variant was estimated to account for 75% of sequence reads in the last two weeks. Though the precise proportion of BA.2.75 at Mt Maunganui is uncertain, this result may reflect an increasing prevalence of this lineage in the region. BA.2.75 was also putatively detected in Tauranga in week 38, and Auckland North Shore in weeks 38 and 39.

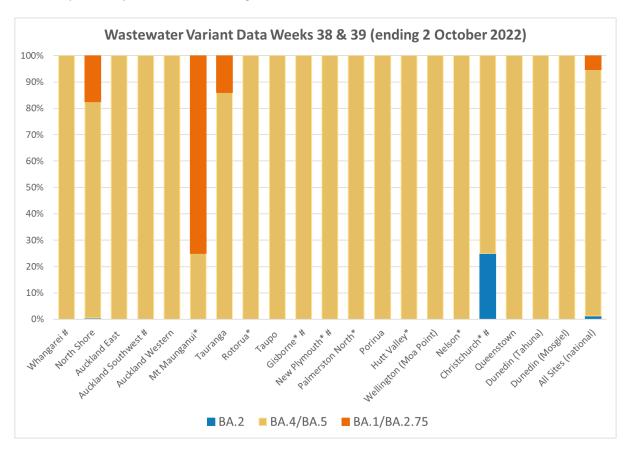


Figure 5. Data from sentinel wastewater sites across NZ using a S-gene (spike) barcoding assay able to 'call' the BA.2 (including BA.2.12.1), BA.4/BA.5 and BA.1/BA.2.75 (sub)variants. Wastewater samples were collected from up to 20 sentinel sites. *The level of precision and sensitivity in these percentage estimates can be uncertain.* Samples marked with an * yielded stochastic detections of one or more variants.

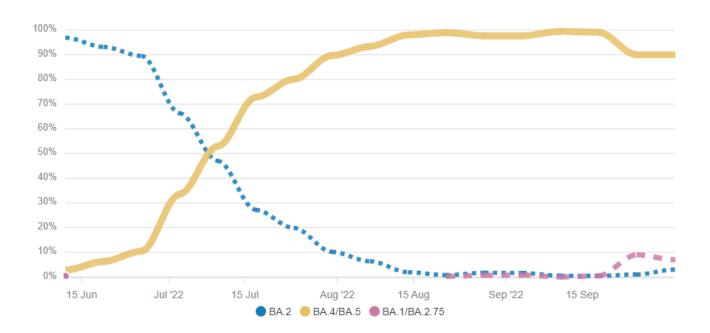
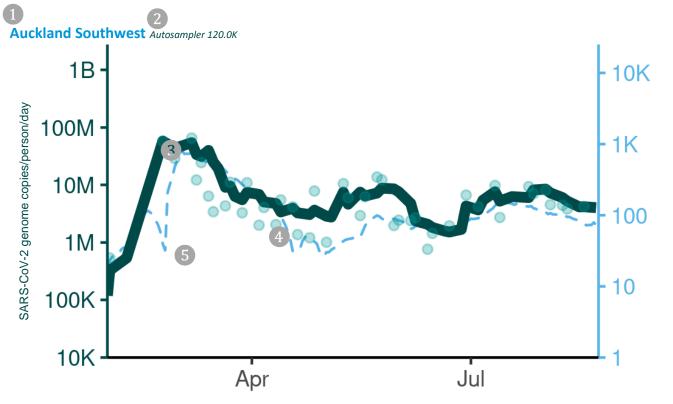


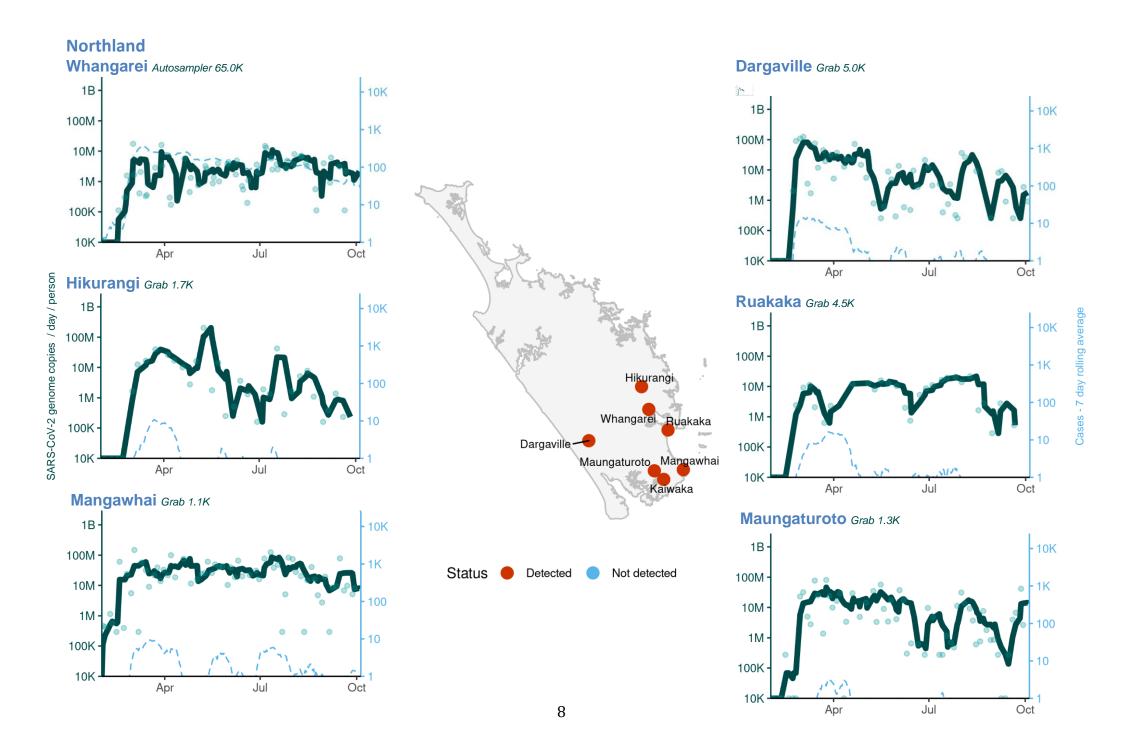
Figure 6. Change in variant prevalence over time at a national scale. Data are collected from up to 20 sentinel sites each week.

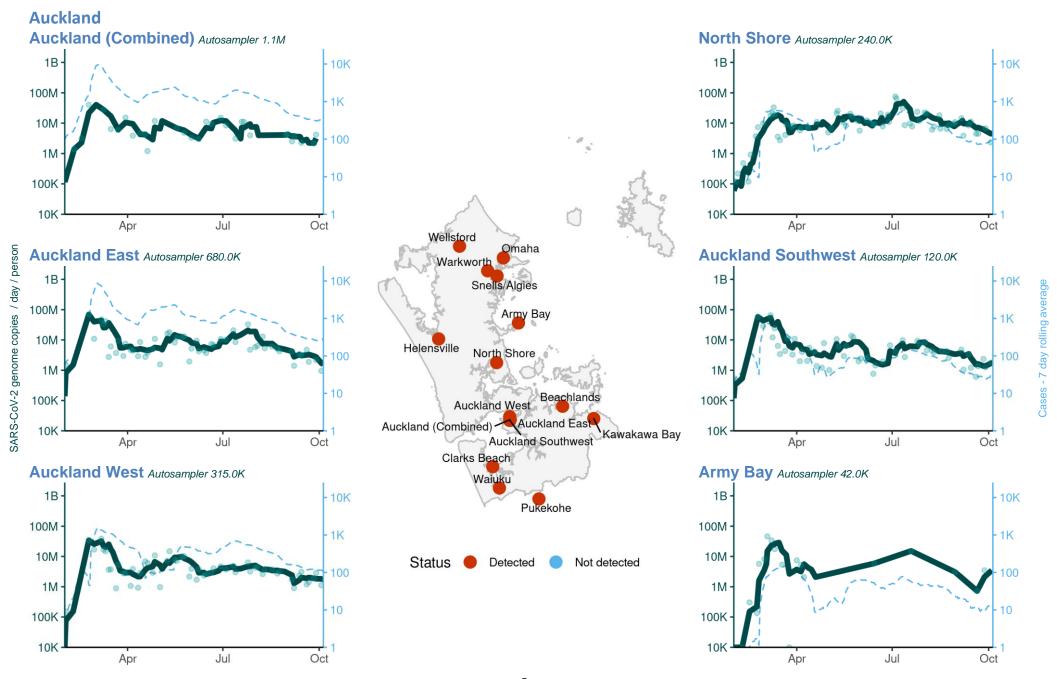
Interpreting Sites Graphs

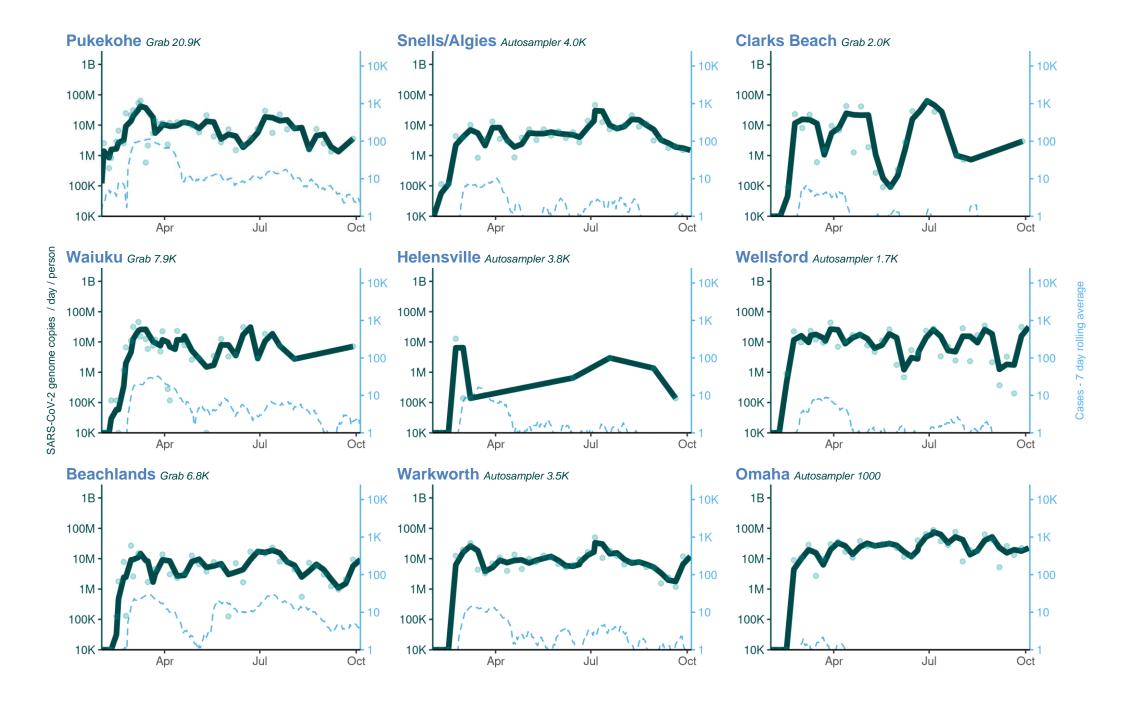


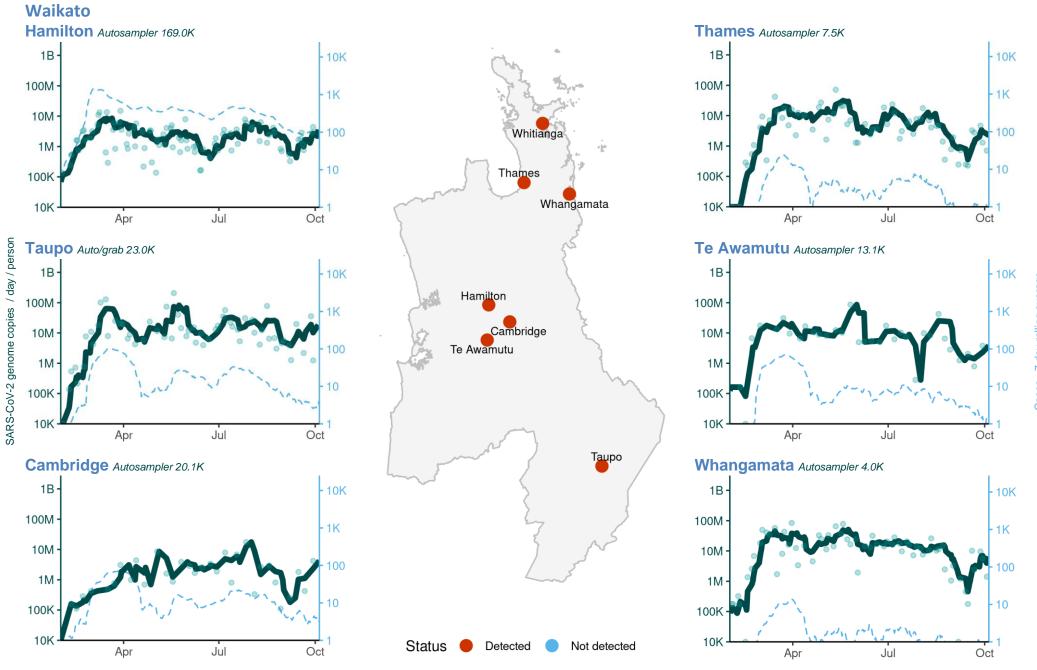
- Site Name
- 2 Sample collection method and population. Results based on autosampler may be more representative than grab sample-based results.
- 3 Wastewater results shown as solid line | 14-day average of genome copies/person/day on a log₁₀ scale.
- 4 Individual results samples shown as circles | Rolling 14-day average of genome copies/person/day on a log₁₀ scale.
- Rolling 7-day average of new cases shown as dashed line | New cases reported in a catchment based on reported date of illness on a log₁₀ scale. This data is not available for all sites and subject to change.

Note: Wastewater and cases data are on a log_{10} scale. Scales on all graphs have been normalized to cover the same scale on every graph. Care should be taken when interpreting the data

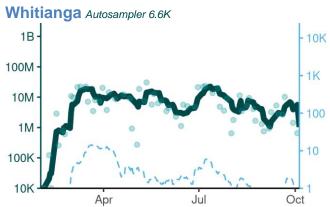


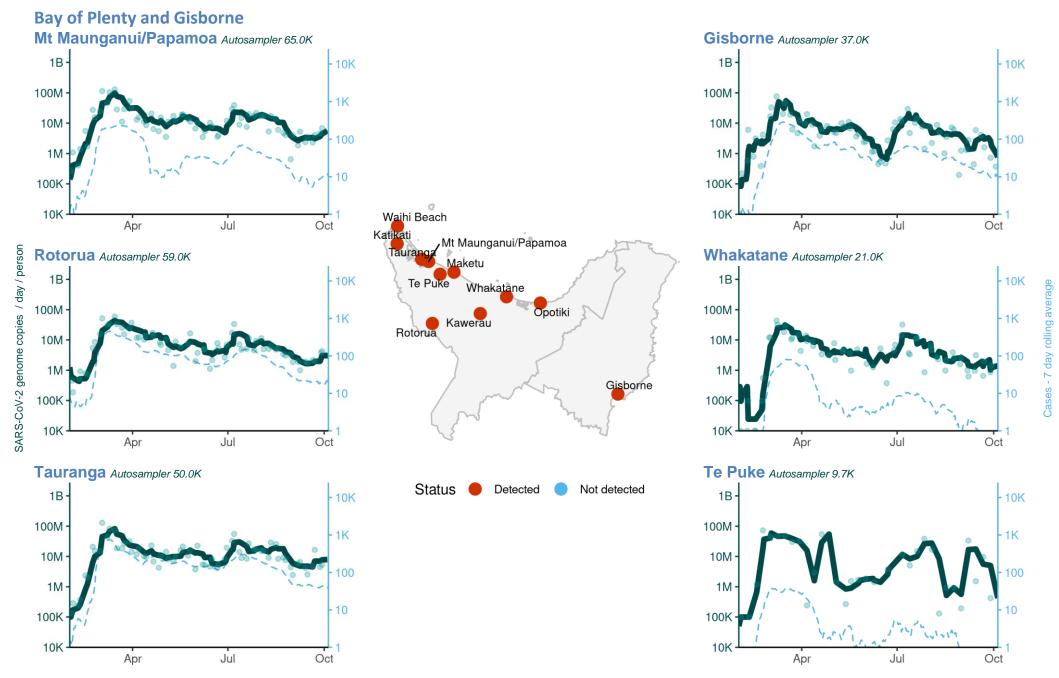


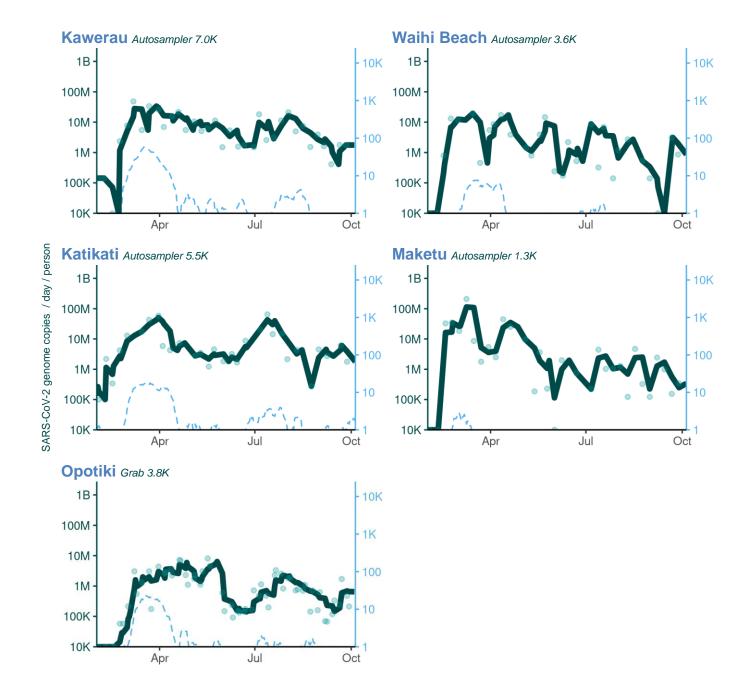


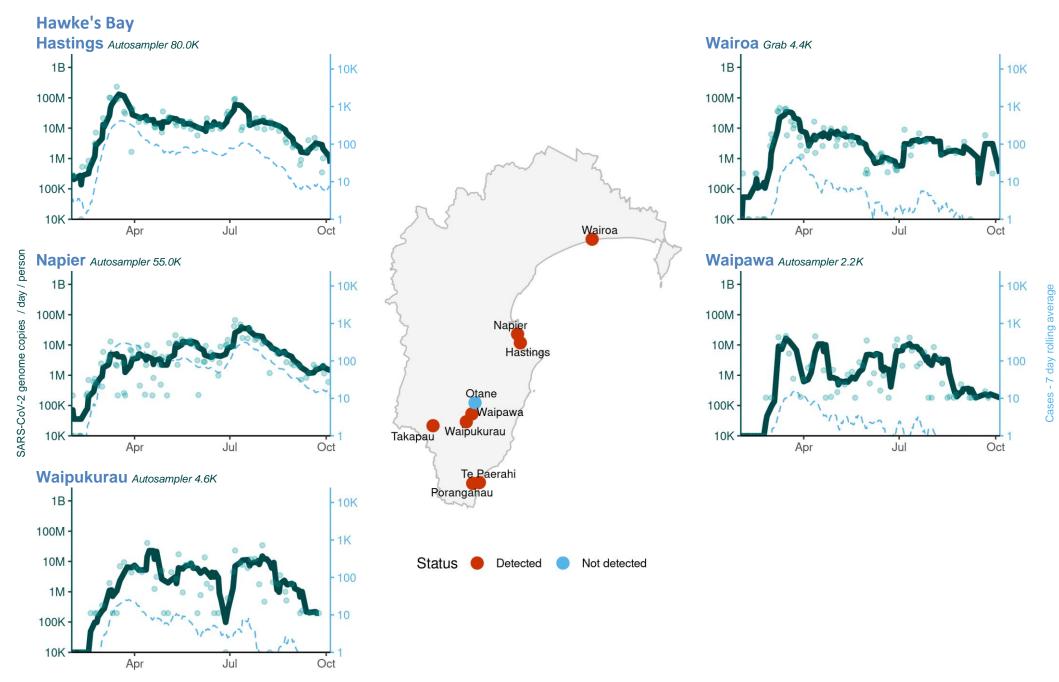


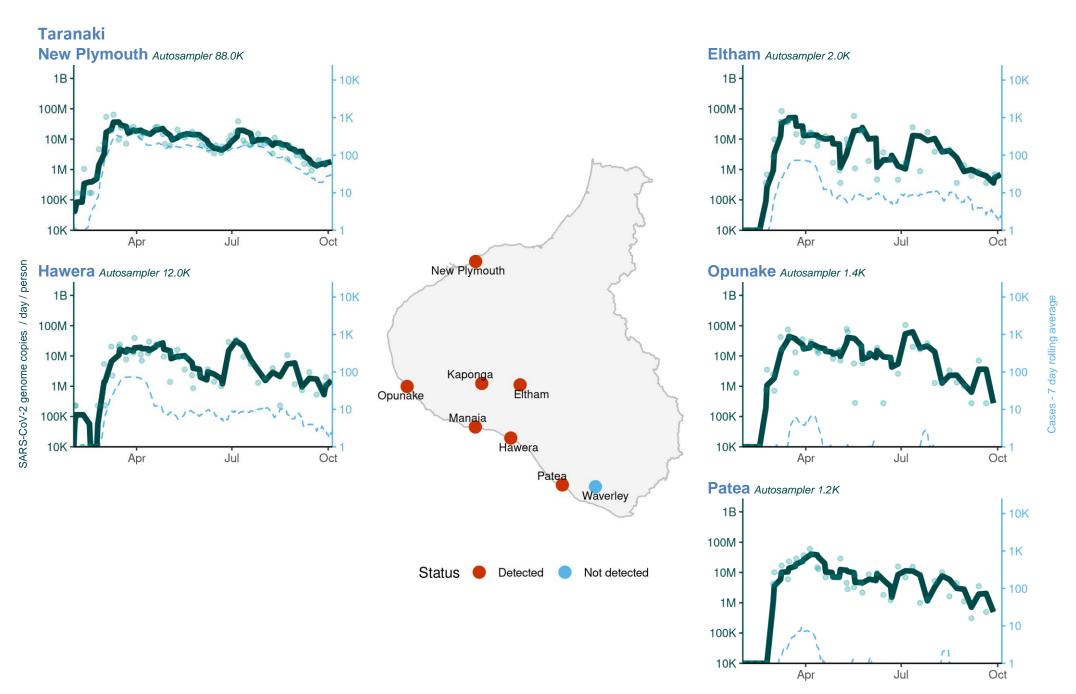


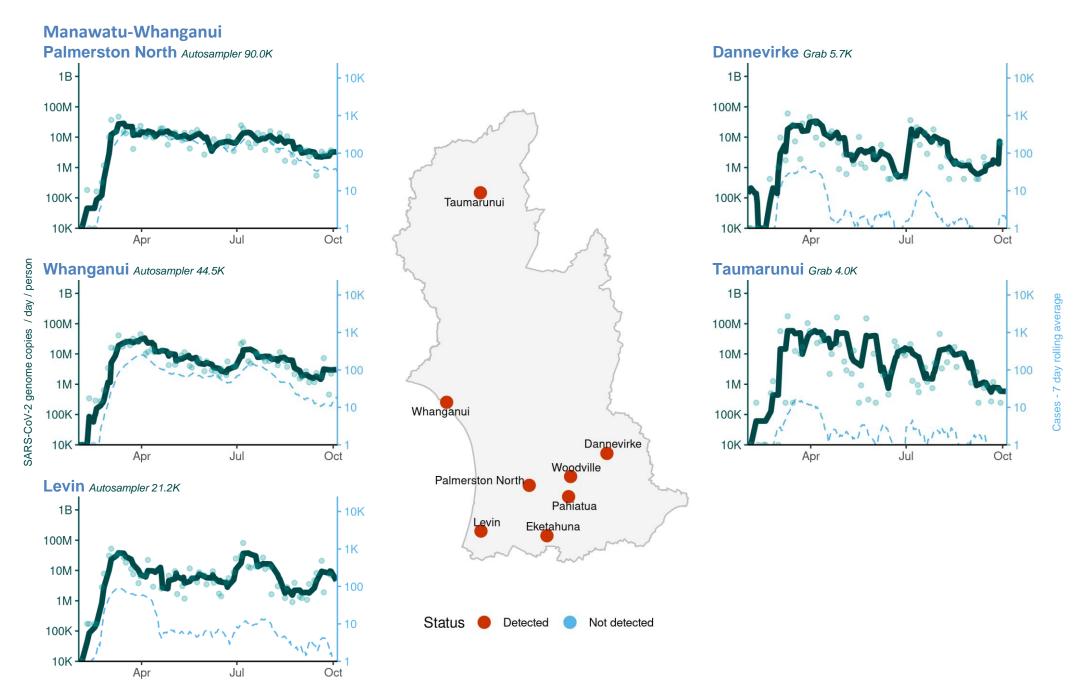


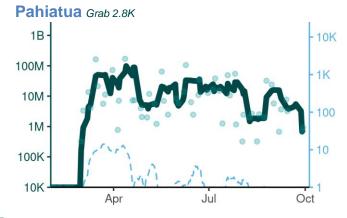


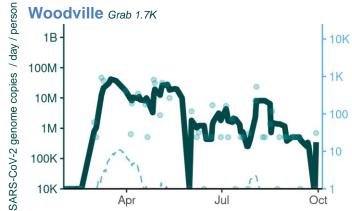


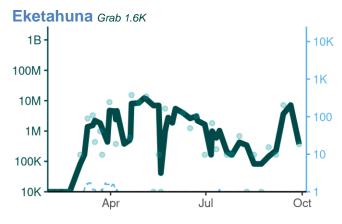


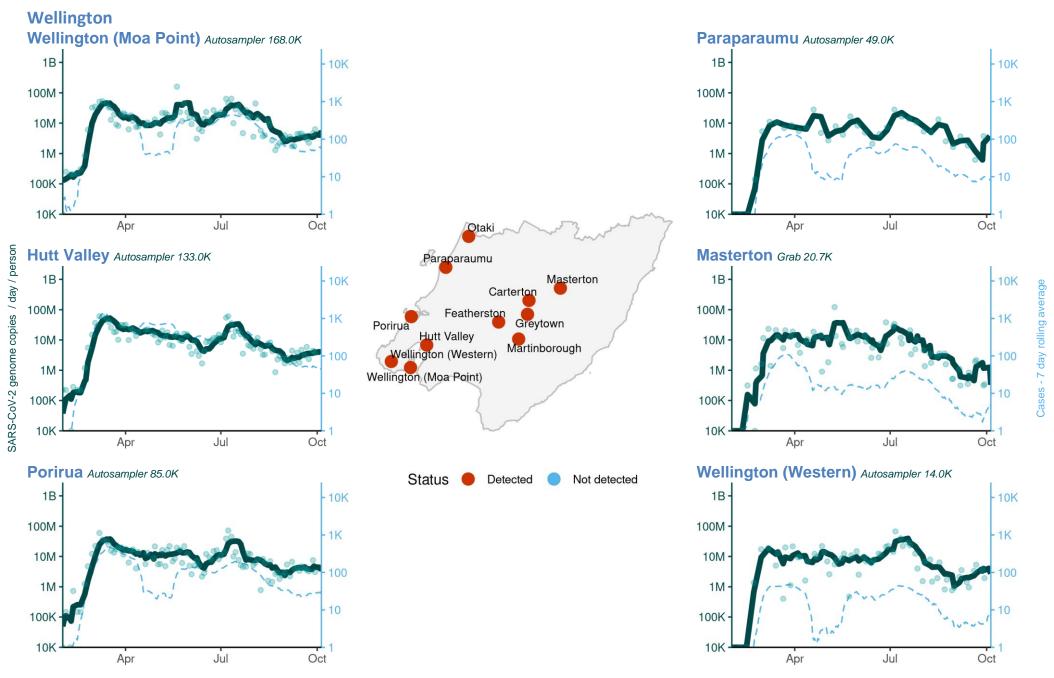


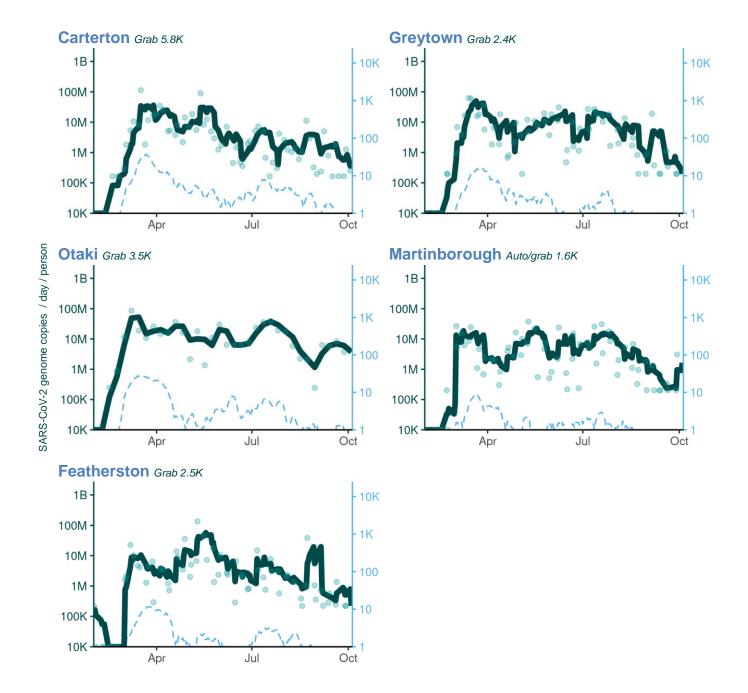


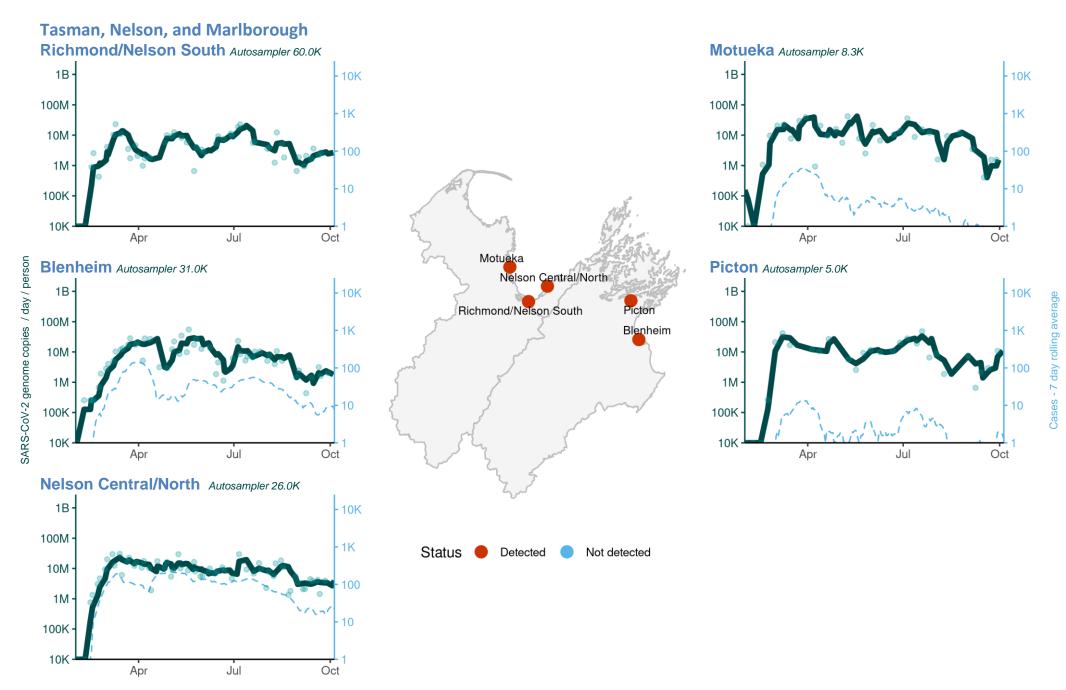


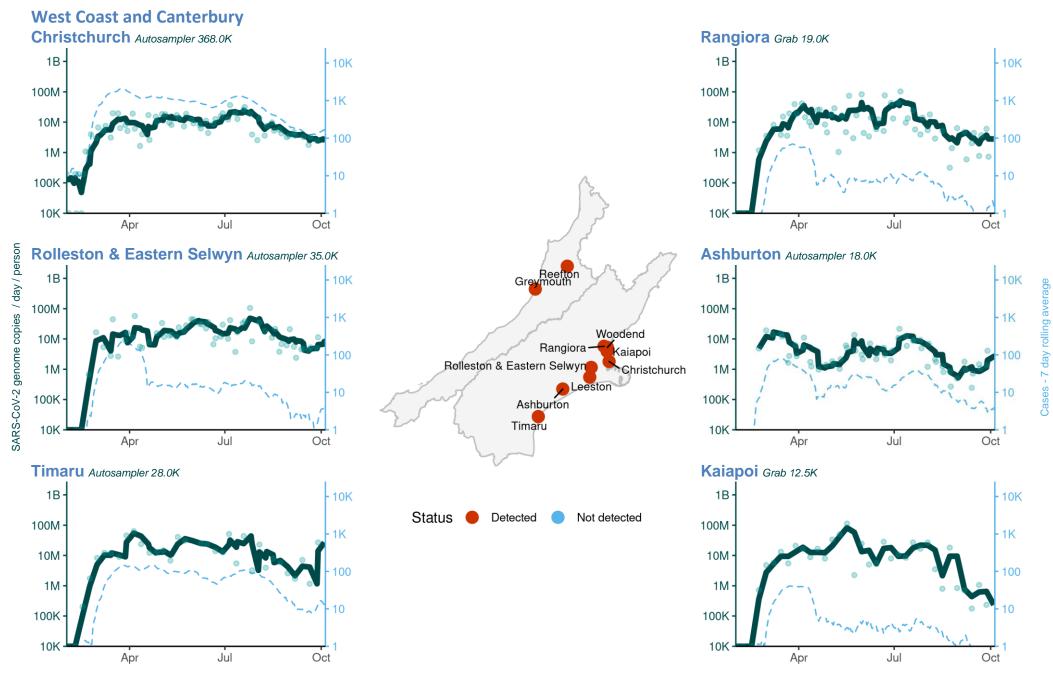












10K

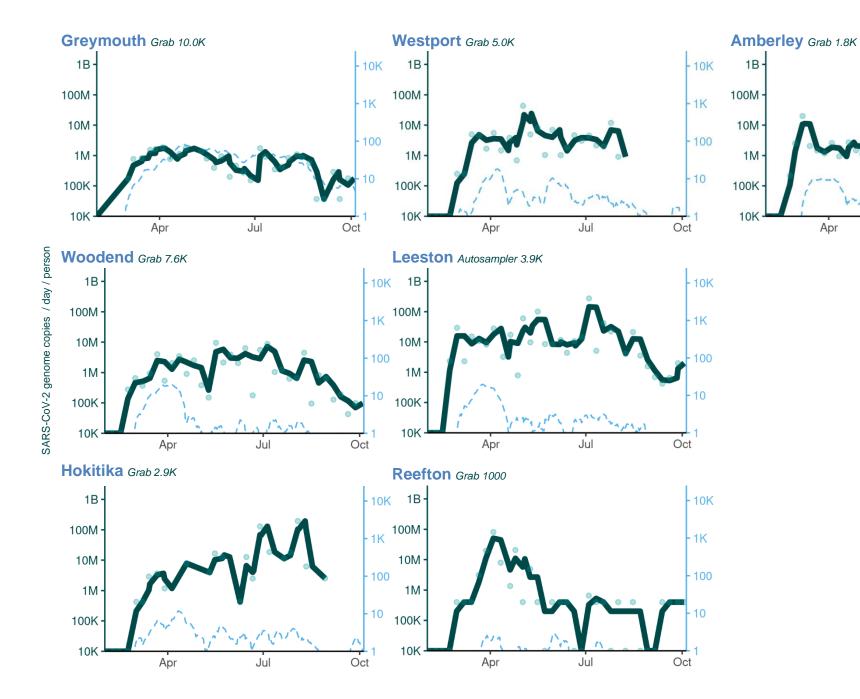
1K

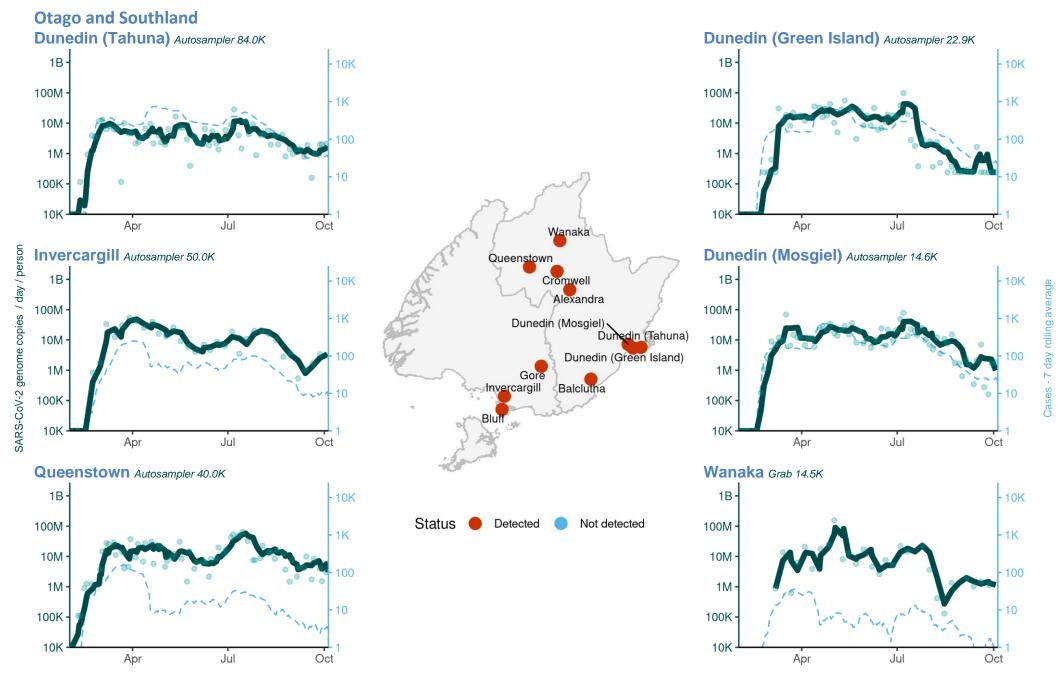
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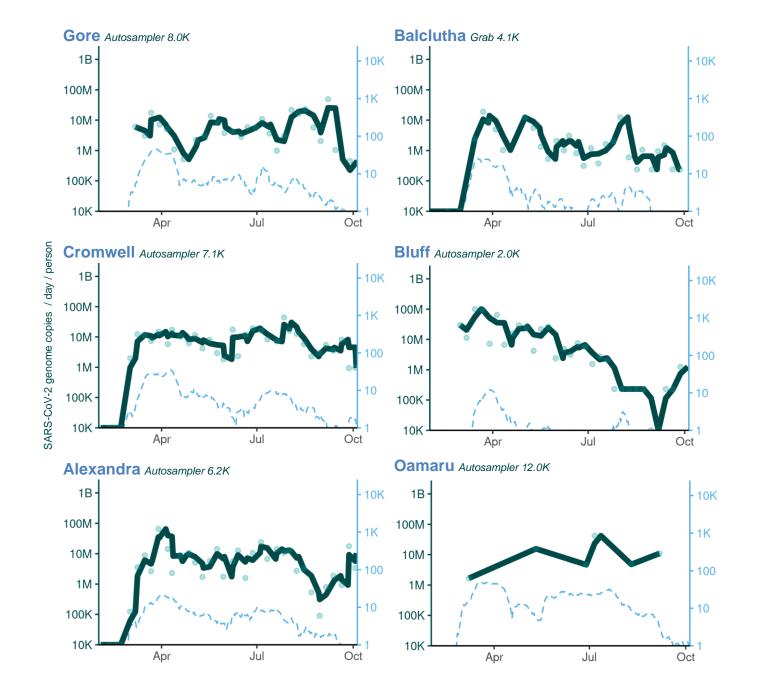
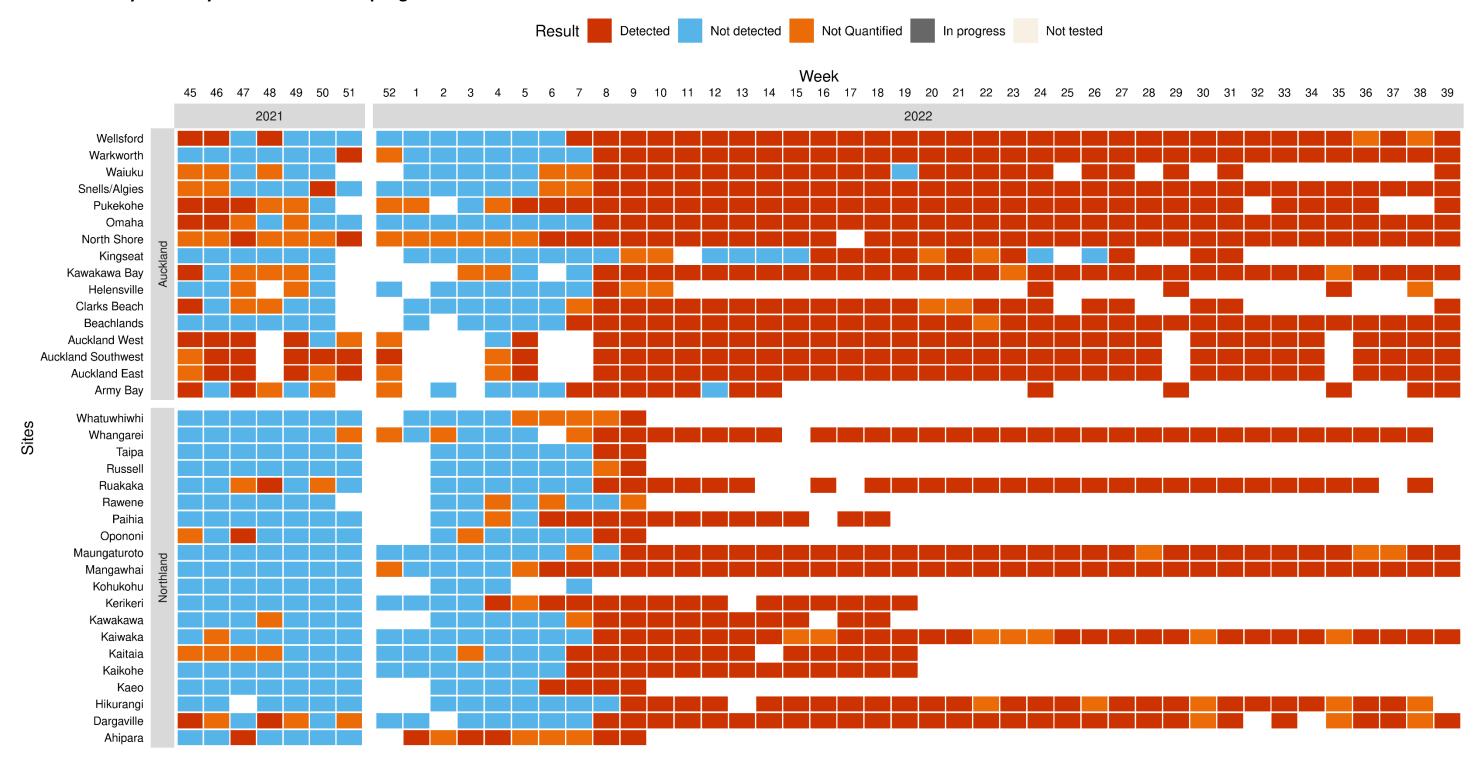
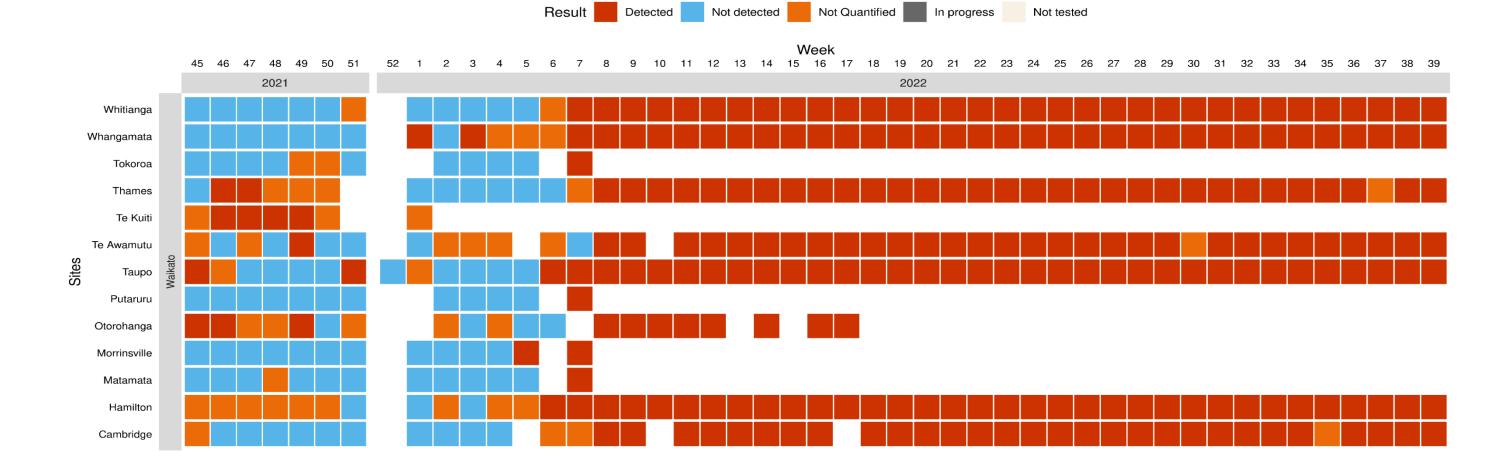
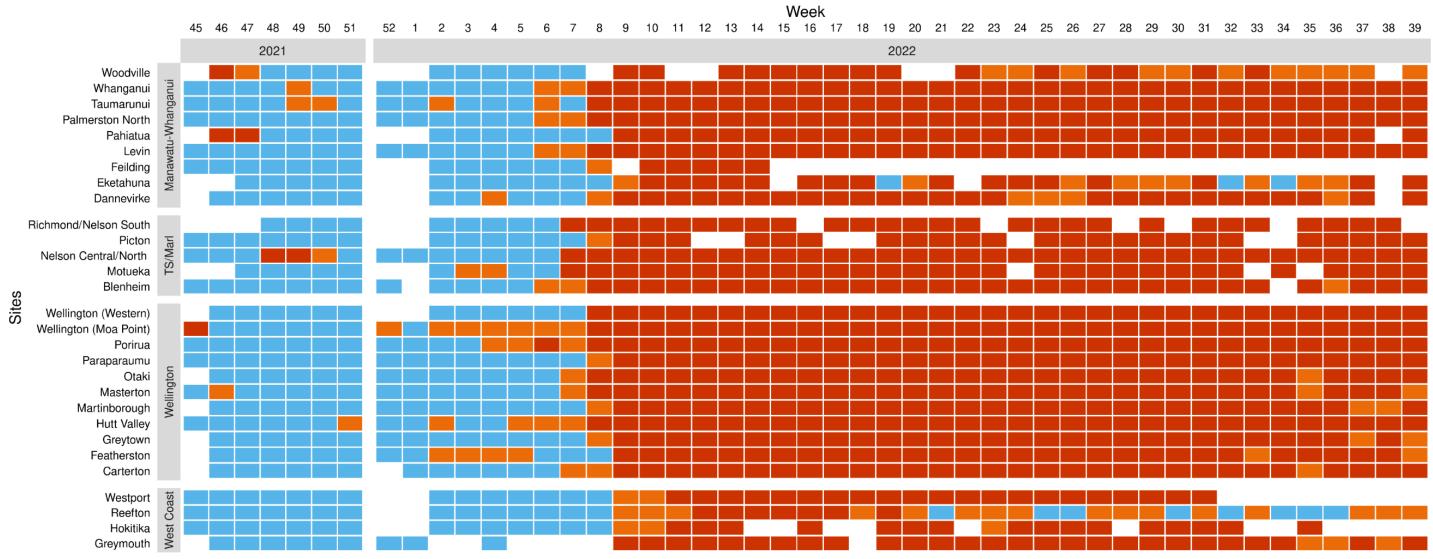


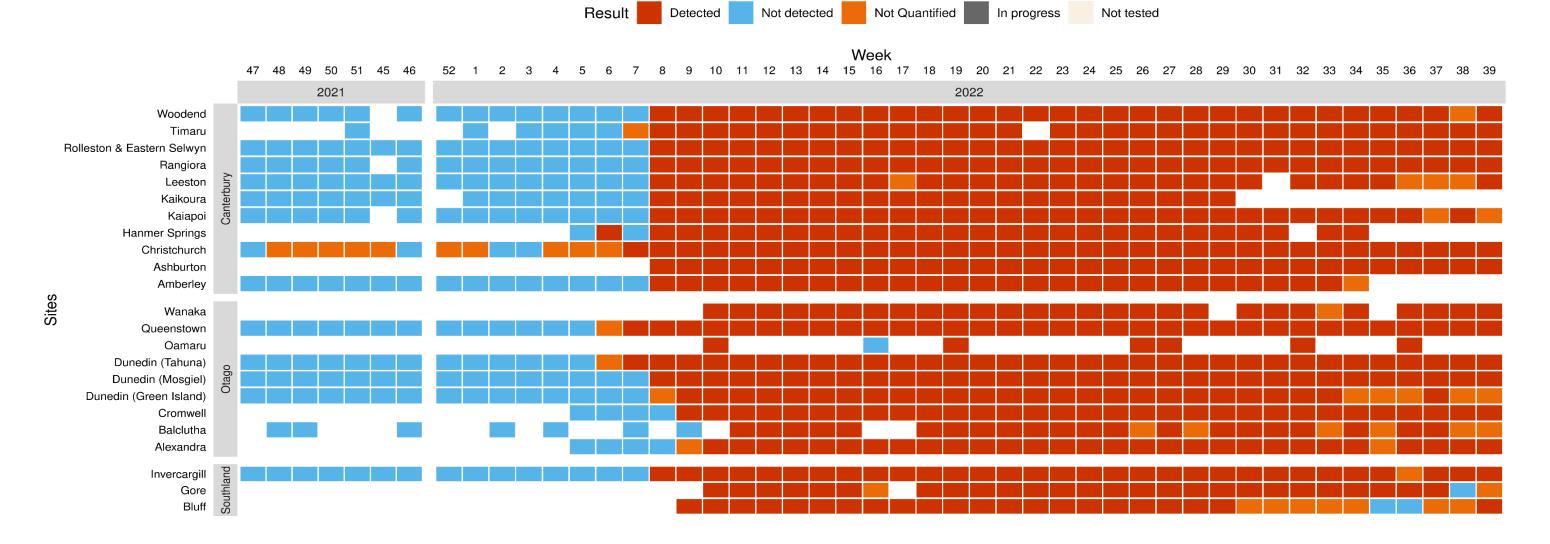
Table 1: Weekly summary of Wastewater sampling results for SARS-CoV-2











Overview

SARS-CoV-2, the virus that causes COVID-19 disease, is shed in the faeces of people that are infected and so the viral RNA can be detected in wastewater. As such, testing wastewater for SARS-CoV-2 RNA is an efficient population-based COVID-19 surveillance tool. Based on national and international data, this method has been shown to be an indicator of increasing and decreasing cases (i.e., early warning system) and complements other surveillance tools. A national wastewater COVID-19 surveillance programme was established in 2021 by the Institute of Environmental Science and Research (ESR). This work is funded by the New Zealand Ministry of Health and is part of New Zealand's COVID-19 response.

Wastewater samples are collected from wastewater treatment plants across both the North and South Island of New Zealand. Grab or 24 hr composite sample s are collected. Most sites are sampled at least weekly between Monday and Thursday of any given week. The number of sites and frequency of collection varies over time. Greater variability is expected with grab samples.

Approach

Samples are sent from each wastewater treatment plant to ESR. Processing involves the concentration of virus and extraction of viral RNA. The presence of SARS-CoV-2 RNA in the sample is then determined using RT-qPCR.

A result of **not detected** means that SARS-CoV-2 RNA is either absent from the sample, or at a level too low to be detected. When SARS-CoV-2 RNA is **detected**, the concentration in the sample can be calculated. Low amounts of SARS-CoV-2 RNA in a sample may not be able to be accurately quantified and are recorded as less than the limit of quantitation. For quantitation, the raw concentration data (i.e., genome copies/L) is converted to a viral load of **genome copies per day per person**. This calculation considers the flow rate of wastewater entering the wastewater treatment plant and the population in the catchment. This is the population-normalised viral load.

Key Points & Limitations

- SARS-CoV-2 RNA concentrations should not be compared between wastewater catchments.
- Day to day variability in SARS-CoV-2 RNA concentrations, especially in smaller catchments, is to be expected. Greater variability is expected with grab samples.
- Generally, increasing viral loads are associated with increasing numbers of people with SARS-CoV-2 infection and vice versa (decreasing concentrations indicating decreasing cases). However, there are a number of factors that affect the amount of viral RNA

- detected and so data from wastewater surveillance cannot indicate the exact number of COVID-19 cases in the catchment area.
- The number of COVID-19 cases reported via individual testing are reported for each region to provide a comparison to the wastewater results. The cases in each catchment area are an estimate of the number of people in that wastewater catchment area that have reported a positive test. However, because the wastewater catchments do not exactly align with regional boundaries, the number of cases estimated by region and by water catchment area may be different.
- Data are provisional and may be subject to change by location.
- As septic tank systems are not connected to wastewater treatment plants, the wastewater from these households will not be represented in the data.

Acknowledgements

This work represents the combined efforts of many individuals and organisations.

We continue to be indebted to the teams across the country who are collecting the wastewater that underpins this work.

The wastewater analysis has been undertaken at ESR by a team which may on any given week include contributions from Joanne Chapman, Dawn Croucher, Joanne Hewitt, Joycelyn Ho, Anower Jabed, Susan Lin, Olivia Macrae, Ashley McDonald, Andrew Ng, Ashley Orton, Paula Scholes and Fatiha Sulthana. Data science analysis, visualisation and reporting is the result of team effort from: Franco Andrews, Bridget Armstrong, Raewyn Campbell, Gerhard de Beer, Richard Dean, Brent Gilpin, Joanne Hewitt, Dawen Li, Helen Morris and Michael Bunce. Ongoing support for this work from the Ministry of Health and ESR management is appreciated.

Notes

Sites and frequency of sample collection: The catchment population sites selected for the surveillance range from approximately 100 to over 1,000,000 individuals. The sites cover all regions of the country. Most major towns and all cities, as well as many smaller communities, are included. In early 2022, the wastewater catchment areas cover over 80% of the population connected to wastewater treatment plants. The sites from which samples have been collected have varied over the last 12 months. New sites may be added over time, and/or sampling may reduce in frequency or cease for other sites. The selection and frequency of sampling vary depending on the local population, access to wastewater collection points, staff availability to collect samples and risk factors. When included, samples are collected at least weekly, with twice weekly sampling being common. A number of samples have also been collected from non-WWTP sites (manholes and pump stations- mostly in Auckland).

Sampling method: The preferred option is to automatically collect a 24 hour 'composite' sample. This is where a pump automatically collects a small volume of wastewater every 15 minutes over 24 hours using a composite sampler. These samplers are available in some wastewater treatment plants. When composite samplers are not available, 'grab' samples are collected. These range from a sample being taken at a single point in time, to 3 samples taken over 30 minutes, to samples collected over a day. Grab samples represent only the composition of the source at that time of collection and may not be as representative as a 24-hour composite sampler. More variation may be expected with grab samples.

Laboratory analysis of wastewater samples: Processing of each sample commences within an hour or two of receipt. Processing involves the concentration of virus from 250 mL sample to approx. 1 mL using centrifugation and polyethylene glycol. Viral RNA is then extracted from a small volume of 0.2 mL concentrate to give a final volume of 0.05 mL The presence of SARS-CoV-2 RNA is determined using RT-qPCR. SARS-CoV-2 is considered detected when any of the RT-qPCR replicates are positive.

RT-qPCR: Reverse transcription (RT) to convert RNA to complementary DNA (cDNA), followed by quantitative PCR (qPCR). RT-qPCR is used for detection and quantification of viral RNA.

Method sensitivity: The protocol used to concentrate SARS-CoV-2 from wastewater allows for the sensitive detection of SARS-CoV-2 by RT-qPCR. ESR has shown that when 10 individuals are actively shedding SARS-CoV-2 RNA in a catchment of 100,000 individuals, there was a high likelihood of detecting viral RNA in wastewater

(https://doi.org/10.1016/j.watres.2021.118032). Shedding by one individual may be detected in wastewater, but it does depend on many factors including the amount and duration of shedding. Very low levels in wastewater may be not able to be quantified (i.e., less than the limit of quantification- see below).

SARS-CoV-2 RNA detected (positive result): A positive detection in the wastewater indicates that at least one person has been shedding SARS-CoV-2 into the wastewater at some point during the time period that the sample was being collected. In some cases, detections could also be due to the shedding of low levels of SARS-CoV-2 RNA by a recently recovered case. The detection of SARS-CoV-2 RNA does not indicate that infectious virus is present.

SARS-CoV-2 RNA not detected (negative result): A negative result can occur because there are no active 'shedding' cases in the catchment or because the SARS-CoV-2 RNA concentration is too low to be detected, most likely because there are a very low number of cases in the wastewater catchment. Therefore, negative finding does not necessarily guarantee the absence of COVID-19 in the community.

Viral loads and normalisation: When detected, the SARS-CoV-2 RNA concentration is calculated as genome copies per L of wastewater. This is then converted to a viral load of **genome**

copies/day/person. This conversion takes into account the flow rate of wastewater entering the treatment plant (the influent) and the population in the catchment. The **flow rate** is the total volume (m3 per day) recorded at the inlet of the wastewater treatment plant over 24 hours. This is a **population-normalised viral load**. Currently, the flow rate is the average annual flow rate, but will be replaced with daily flow rate when available (note that rainfall may significantly increase the flow rate at the inlet, diluting the sample, and may result in lower concentrations and a false negative result).

In future, SARS-CoV-2 RNA concentrations will also be normalised by testing for the presence of pepper mild mottled virus (PMMoV). PMMoV is a virus that infects peppers but not humans. Consumption of peppers or pepper products, such as chilli sauce, means that PMMoV is detected in wastewater – normally at very high concentrations. Therefore, PMMoV has been found to be a useful proxy for the amount of faecal material in a wastewater sample. For normalisation, the concentration of SARS-CoV-2 RNA is divided by that of PMMoV in each sample. Different normalisation methods may result in changes to some data points, but trends are unlikely to change significantly.

Limit of quantification: The lowest concentration of the target that can be reliably quantified is referred to as the limit of quantification. For those samples where SARS-CoV-2 is detected but cannot be quantified, a value of 5 genome copies/mL wastewater is used. While a standard method is being used, virus recovery can vary from sample to sample, and this may affect the quantitation.

Data subject to change: Data generated for the New Zealand Wastewater COVID-19 Surveillance Programme should be considered provisional and may be subject to change. Data may be incomplete for the most recent 2-week period due to processing, testing and reporting delays.

Data not shown:

- Data from 'ad hoc' sampling locations including from individual facilities/building (e.g., workplaces, prisons, MIQs) are not included.
- Results from certain samples may not be shown, as the result was either deemed invalid, or the sample could not be tested (e.g., leaked in transit, not labelled).

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