Grid monitoring of SARS-CoV-2 in sewage for an early-warning sign of community outbreak

Tong Zhang

On behalf of the whole team

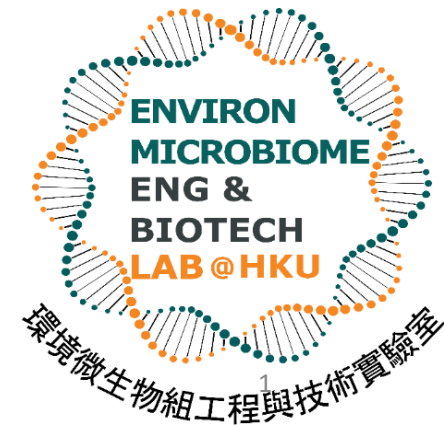Environmental Microbiome Engineering and Biotechnology

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Civil Engineering: T Zhang, Y Deng, XW Zheng, XQ Xu, SX Li, JH Ding
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Government:
FHB:
        HMRF office: The financial support from Health and Medical Research Fund
        CHP: Sharing clinical/epidemiological data
HA: Sharing clinical/epidemiological data
Environment Bureau:
        EPD Team :   Strategic planning and catchment analysis
        DSD Team :   Sewer network analysis and sampling arrangements

Wastewater-based Epidemiology
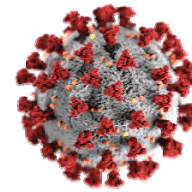
Daughton (2001)
Propose the idea of **community-scale surveillance** to monitor the usage of illegal drugs in USA.

Daughton (2011), Fanelli (2011), Thomas and Reid (2011)
"real-time monitoring of sewage for biomarkers as measures of **community-wide health and disease**".

Gertjan Medema (March, 2020)
Use sewage surveillance to monitor community circulation of SARS-CoV-2 virus in Netherlands.

SARS-CoV-2 outbreak in 2020

Zuccato et al. (2005)
Applied **wastewater analysis** to estimate community illegal drug consumption in Italy.

Daughton (2012),
Put forward the concept-**BioSCIM** (sewage chemical information mining targeted at biomarkers) and **SCIM** (sewage chemical-information mining).

WHO (August, 2020)
Recognize wastewater surveillance of SARS-CoV-2 as a potentially useful tool to monitor community circulation.

"Wastewater-Based Epidemiology (WBE) is a new epidemiology tool that has potential to act as a complementary approach for current infectious disease surveillance systems and an early warning system for disease outbreaks."
(https://www.sciencedirect.com/science/article/pii/S0160412020304542?via%3Dihub)

SARS-CoV-2 in wastewater provides valuable data

THE LANCET
Gastroenterology & Hepatology

CORRESPONDENCE | ONLINE FIRST

SARS-CoV-2 in wastewater: potential health risk, but also data source

Willemijn Lodder • Ana Maria de Roda Husman ✉

nature

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How sewage could reveal true scale of coronavirus outbreak

Wastewater testing could also be used as an early-warning sign if the virus returns.

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SARS-CoV-2 in Feces of Patients

Fecal positivity rate ranges from 15.3% to 100%, with an average value of 48.1%.

Additionally, the patients after negative respiratory system samples may still shed virus in their feces.

Stool virus loading varies from 2~8 log 10 copies/mL with the average value of 4.5 log 10 copies/mL.

(https://www.nature.com/articles/s41575-021-00416-6)

Summary of patients with SARS-CoV-2-positive fecal or rectal swabs.

Author	Region	Number of patients with positive stool/rectal swab sample	Duration of positive infection (days)	Patients with positive stool/ rectal sample after negative respiratory system samples
Adult patients				
Lin et al.[72]	Guangzhou, China	46/217 (21.2%)	3–18	30/46 (65.2%)
Ling et al.[73]	Shanghai, China	54/66 (81.8%)	9–16[a]	43/55 (78.2%)
Cheung et al.[15]	Hong Kong, China	9/59 (15.3%)	Data collection on presentation	NA
Kujawski et al.[74]	USA	7/12 (58.3%)	1–12	1/7 (14.3%)
Lo et al.[75]	Macau, China	9/9 (100%)	1–18	1/9 (11.1%)
Young et al.[76]	Singapore	4/8 (50%)	1–7	1/4 (25%)

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Why Sewage Test?

- From US CDC : "Data from wastewater testing are not meant to replace existing COVID-19 surveillance systems, but are meant to complement them by providing:
  - An efficient pooled community sample.
  - Data for communities where timely COVID-19 clinical testing is underutilized or unavailable."

https://www.cdc.gov/coronavirus/2019-ncov/cases-updates/wastewater-surveillance.html

- Patients not willing to take clinical tests due to different reasons.

- Patients have no symptom.

- Non-invasive surveillance compared to some other alternatives.
  (https://www.nature.com/articles/d41586-020-00973-x)

Applications of Sewage Surveillance of SARS-CoV-2

✓ Provide early signals for a catchment area

✓ Identify the local hotspot buildings/area

✓ Monitor trends in community transmission

✓ Assess viral removal in WWTPs

✓ Trace viral origins by sequencing or AS-RT-qPCR

A study done by MIT

Eric J. Alm

Professor of Civil and Environmental Engineering and Biological Engineering

Associate Member, Broad Institute

High-quality correlation of case *vs* virus detection data

- 116 sewage samples in the entire outbreak period from middle February to May 2020.

- They concluded that the dynamics of SARS-CoV-2 in sewage treatment works can be used to foreshadow the trend of COVID-19 transmission, with **4-10 days** in advance of clinical cases reporting.
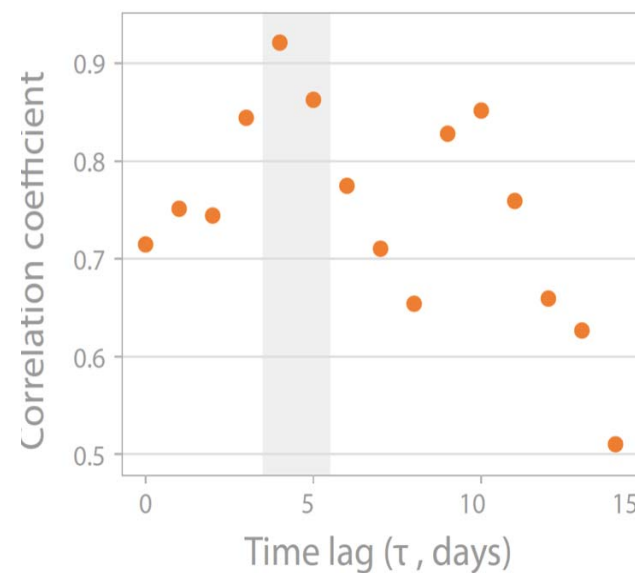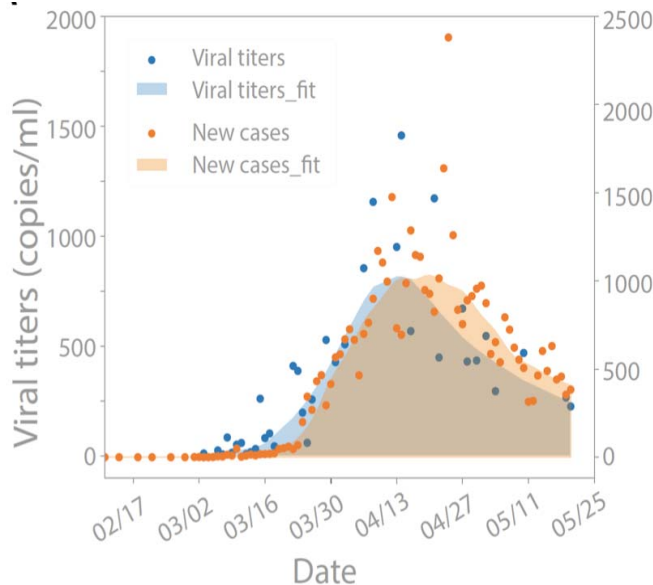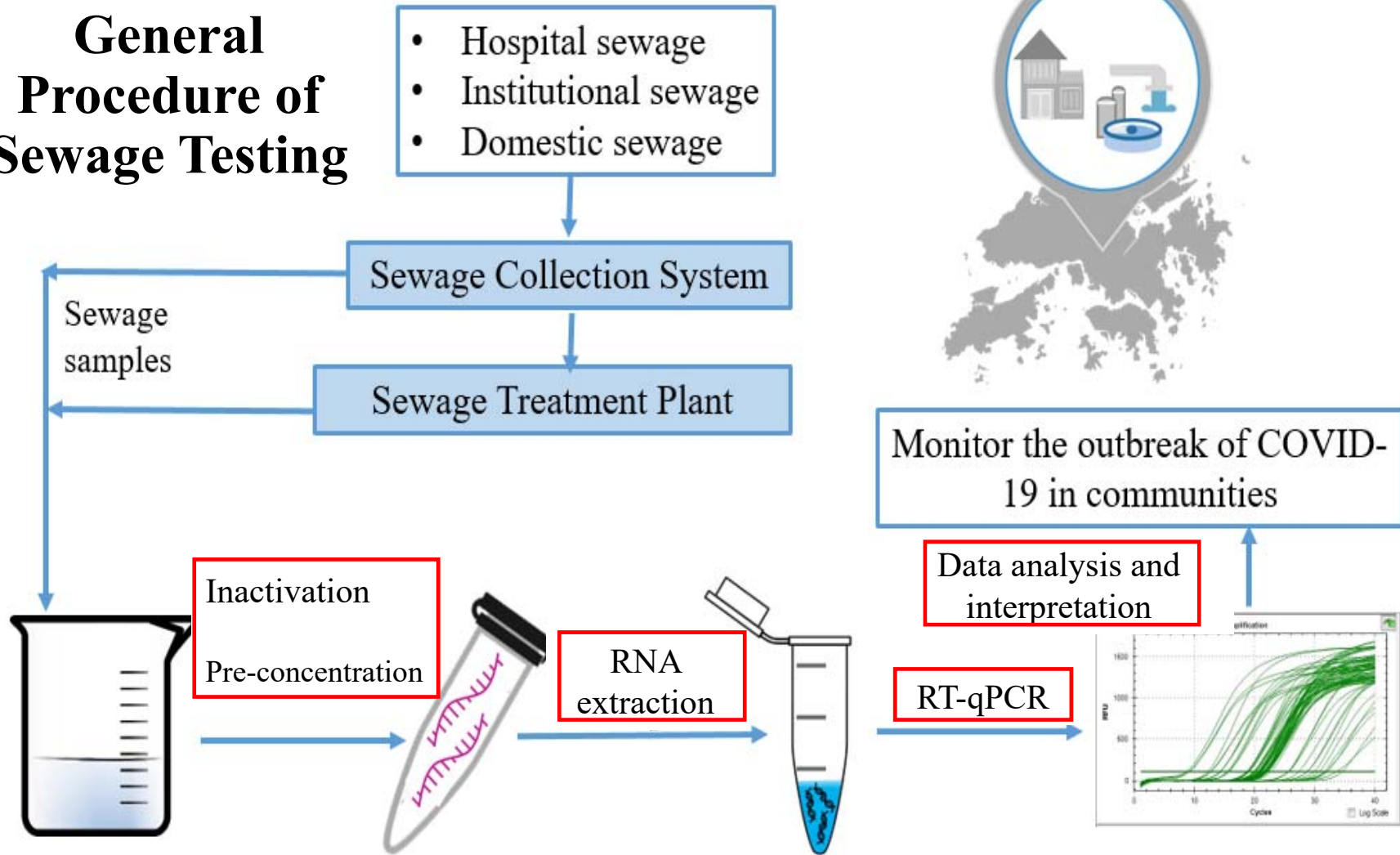
Viral titers correlate with daily new cases with **a 4-day time lag**. (from MIT *medRxiv, https://www.medrxiv.org/content/10.1101/2020.06.15.20117747v2*)

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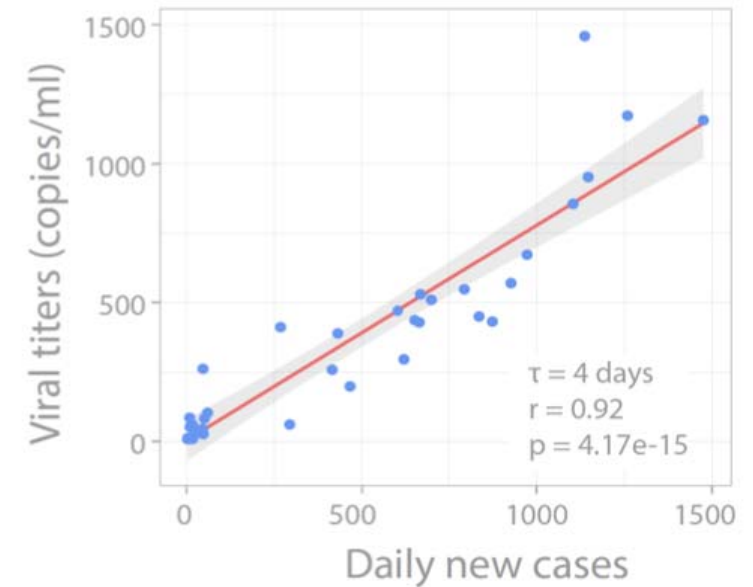
Some oversea projects

- Australia in May of 2020 said it intended to roll out a big project of raw sewage testing for virus in wastewater, so it could focus testing and tracing neighborhoods with problems.

- https://www.scmp.com/week-asia/health-environment/article/3088466/singapore-checking-peoples-poo-coronavirus 2020/06/10 **Singapore**

- https://www.cranfield.ac.uk/press/news-2020/work-begins-on-uk-system-to-detect-covid-19-in-wastewater: 2020/07/16 National COVID-19 Wastewater Epidemiology Surveillance Programme (N-WESP), led by the **UK** Centre for Ecology & Hydrology (UKCEH)

- https://www.cnbc.com/2020/09/25/hhs-wants-to-test-30percent-of-us-wastewater-for-the-coronavirus.html 2020/09/25 **US Department of Health and Human Services**

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General Procedure of Sewage Testing

- Hospital sewage
- Institutional sewage
- Domestic sewage

Sewage Collection System

Sewage samples

Sewage Treatment Plant

Monitor the outbreak of COVID-19 in communities

Inactivation

Pre-concentration

RNA extraction

RT-qPCR

Data analysis and interpretation

HMRF (Health and Medical Research Fund) project of HKU

- To provide early warning signals for hotspot communities and/or target populations.
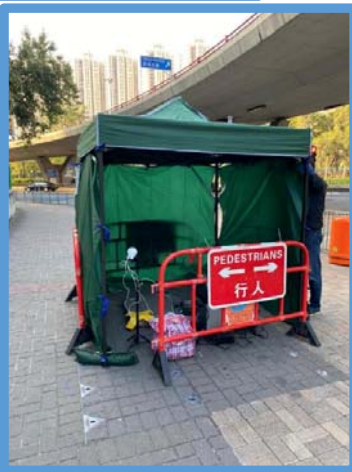- To monitor the trend of outbreak (increase) and subsidence (decrease) of COVID-19.

We designed the sampling plan
1) to focus on those areas with more cases in the 3rd wave;

2) based on intensive discussions among HKU team, ENB/EPD/DSD, FHB/CHP, etc.;

3) considering the sampling feasibility of the sites. The sampling feasibility was evaluated by site visits of DSD team and HKU team.
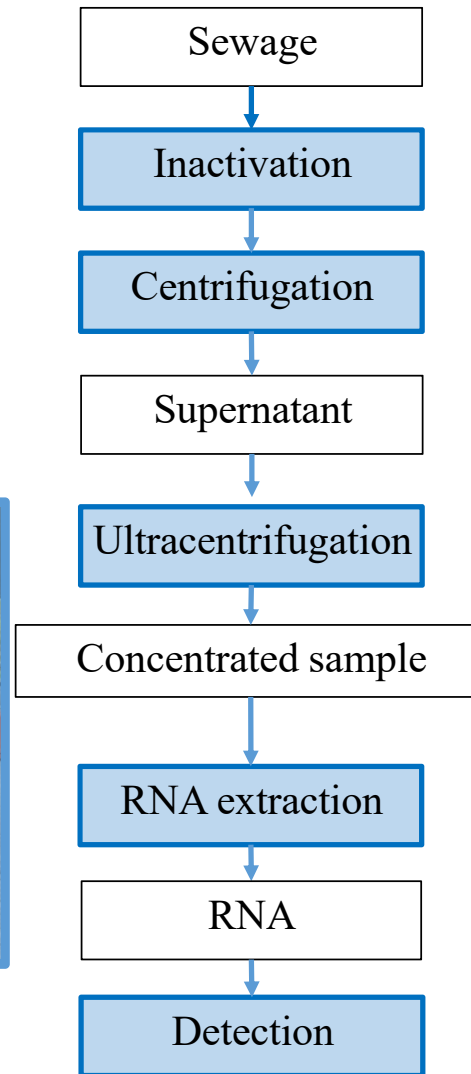
Our Practices for Sampling

- Sites
    1. Manholes of one individual building or a cluster of buildings
    2. Sewage treatment facilities (sewage pumping station, sewage screening plant, and sewage treatment works)

- Frequency (depends on the sampling capacity and purpose of sewage tests)
    Daily, Semiweekly (twice in a week), Weekly, Biweekly

- Type of samples (composite sample *vs* grab sample)
    1. 3 hours in the morning session for manholes under the building
    2. 12 hours at pumping station
    3. 24 hours at wastewater treatment plant

- Safety
    1. PPE wearing during sampling
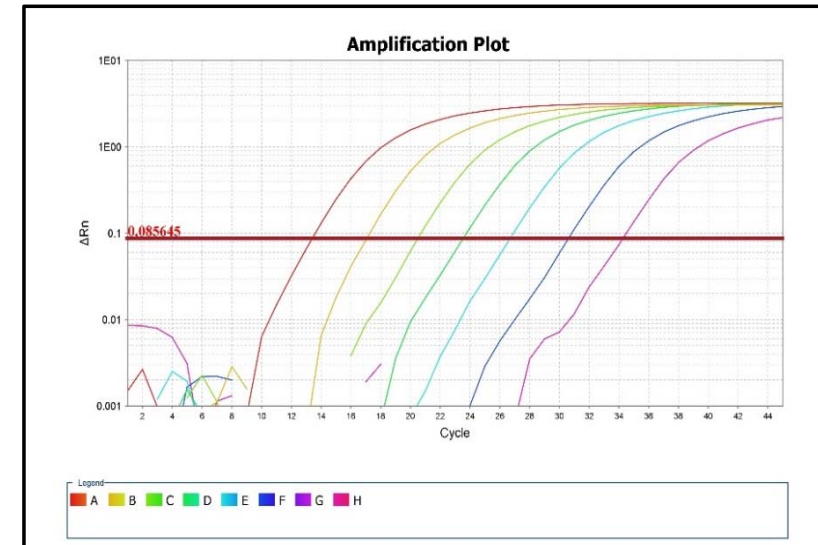   2. Disinfections at the sampling sites after sampling

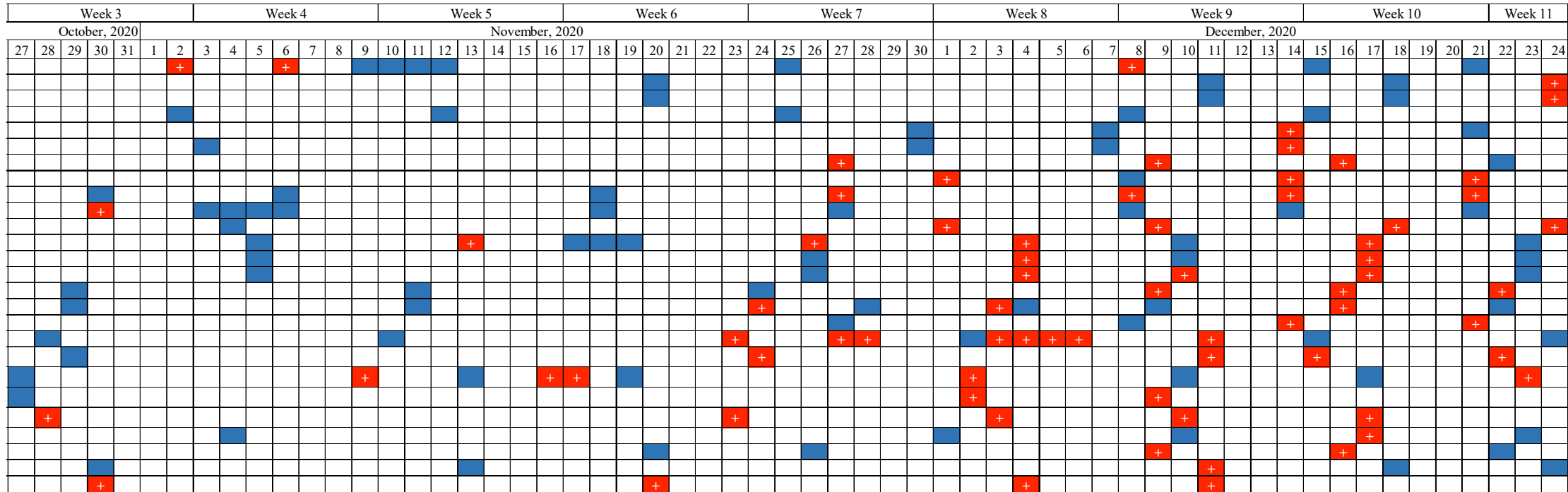# Sampling and Laboratory analysis

Sampling by DSD team

```
Sewage
  ↓
Inactivation
  ↓
Centrifugation
  ↓
Supernatant
  ↓
Ultracentrifugation        Detection
  ↓                        RT-qPCR
Concentrated sample
  ↓
RNA extraction
  ↓
RNA
  ↓
Detection
```

Results for 26 stationary sites during 2020-10-13 to 2020-12-24

Tracking the development trend of community outbreak

Number of cases

Compulsory testing triggered by sewage tests at buildings without previous cases

2020-12-28   **Compulsory test notice** issued to **Fung Chak House** due to consecutive positive signals of sewage tests.

2020-12-29   **3** preliminary positive cases found in Fung Chak House

2020-12-30   Compulsory test notice issued to **Kai Fai House (another building)** based on consecutive positive signals of sewage tests.

2020-12-31   **1** preliminary positive cases found in Kai Fai House

2021-01-02   **1** more preliminary positive case from Fung Chak House

2021-01-06   CTN issued to Fung Chak House for **the second time** due to consecutive positive signals of sewage tests.

2021-01-08   **3** preliminary positive cases from Fung Chak House

2021-01-09   **1** more preliminary positive cases from Fung Chak House

Providing early warning signals for COVID-19 outbreak and monitoring the status of estates with infection clusters

Restriction-Testing Declaration for Restricted Area

Overall, 90% percent of the tested sewage samples were "positive".

Order triggered by sewage testing for compulsory testing (Jan. 23- 24, 2021)

13 hidden cases were identified during Restriction-Testing Declaration Period in city blocks, which are highly correlated with positive sewage testing results.

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Sewage testing uncovered Delta variant in community sewage

- Allele-specific RT-qPCR with primer-probe sets for multiple unique and specific mutation sites of different types of variants.
- Identify the type of variants with a high level of accuracy.
- Shorter turn-over time (a few hours).
- Useful information for quick follow-up actions.

- Successfully identified the Delta variant in a sewage sample collected on June 21, 2021 from Tai Po.
- Alerted the first introduction of the Delta variant in local community sewage

Discovery of Delta variant in Tai Po a 'red alert'

2021-06-24 HKT 18:53

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Discovery of Delta variant in Tai Po a 'red alert'

Implementation of our research in Hong Kong

> 1,500 sampling sites in total

> 5.3 million people (70% of total population in Hong Kong)

- Daily monitoring for community sewage since October 2020.

- Once a sewage sample "positive" for SARS-CoV-2, the health authority will consider compulsory testing to find the hidden COVID-19 patient.

HKU DSD EPD FHB ENB
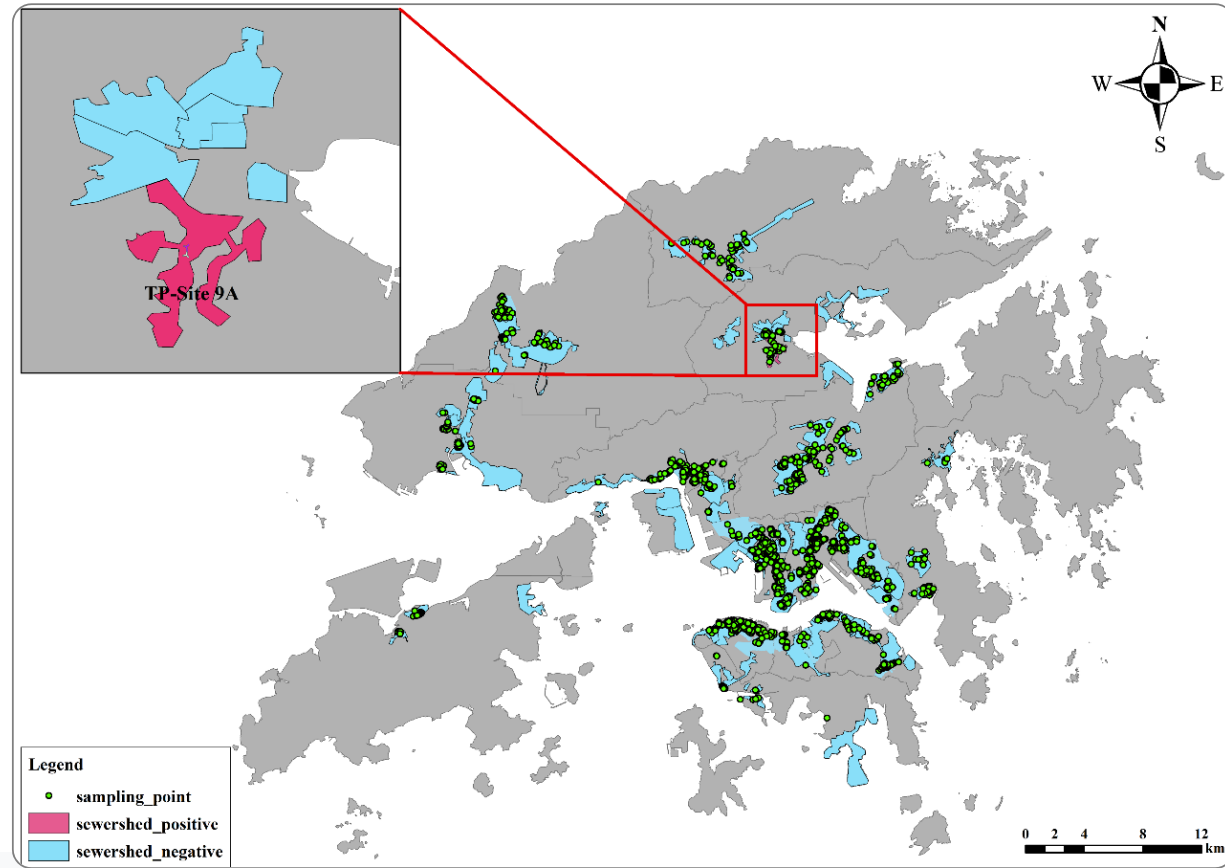
The city-wide sewage surveillance system in Hong Kong

Take-home messages

- The sewage monitoring system has identified more than 50 confirmed COVID-19 cases in Hong Kong so far.

- The established the routine sewage monitoring system, and now covers over 100 regular sampling sites, providing early warning signals of COVID-19 re-emergence for over five million people.

- The experience on COVID-19 sewage surveillance will help build up substantial and long-term collaborations between wastewater specialists, epidemiologists, government decision makers and even the private sectors to explore the opportunities of mining wastewater as an important information source of public health in a city.

"Sewage may tell the health of a city, and we are learning how to listen to it."