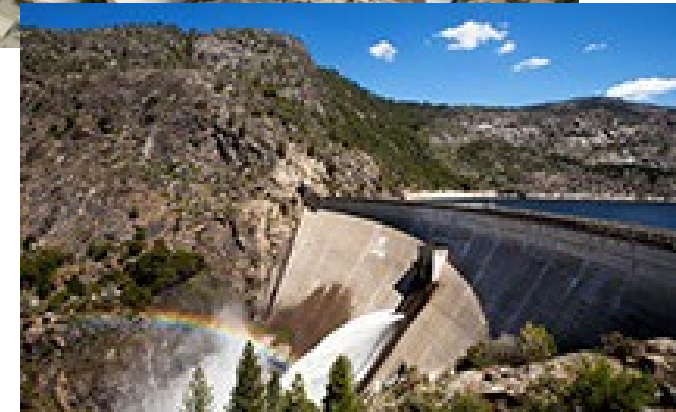




**SFPUC wastewater surveillance
for SARS-CoV-2**

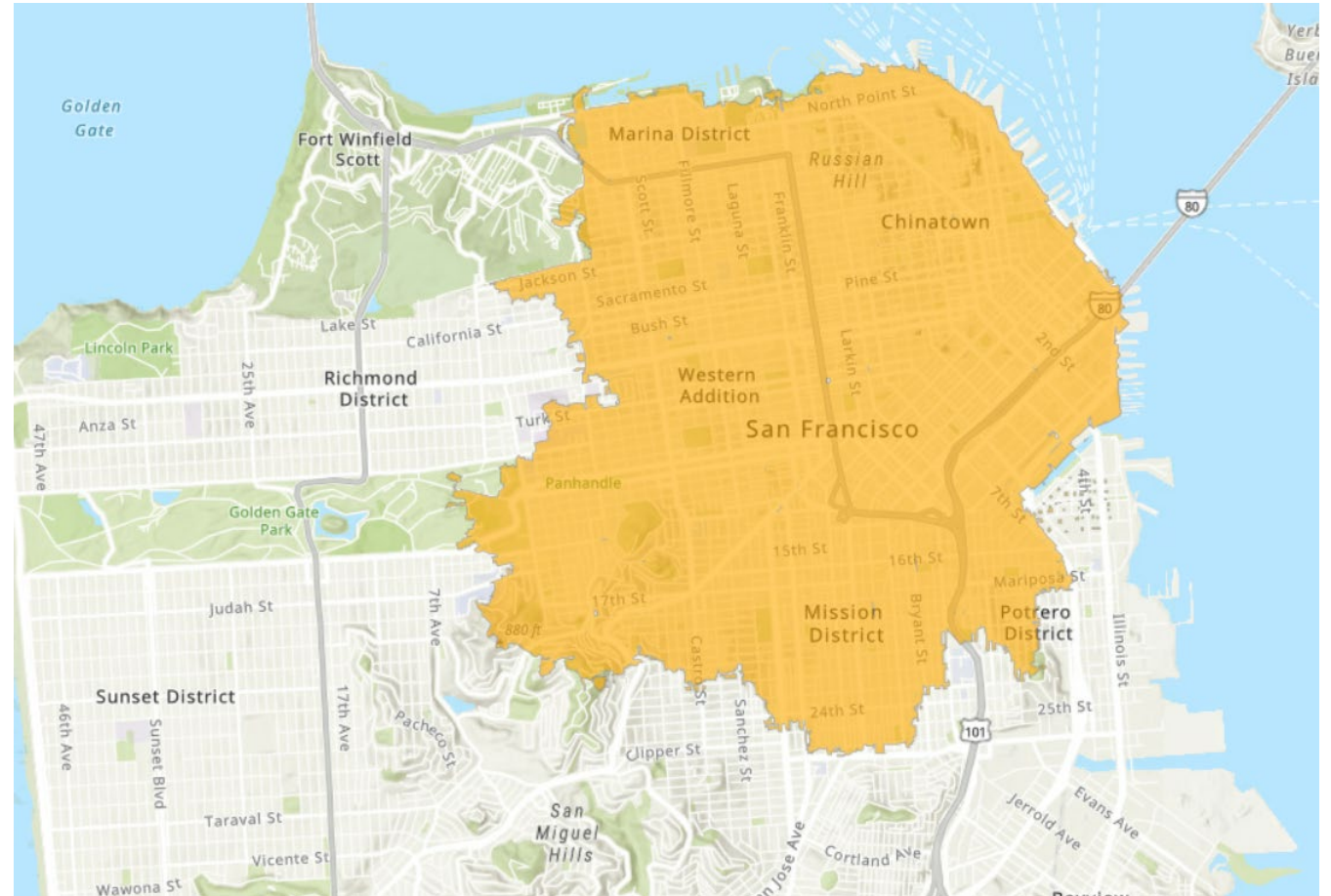
San Francisco Public Utilities Commission

- Water to the city of San Francisco and 3 other counties
- Wastewater management in San Francisco
- Clean power for more than 70% of electricity consumption in San Francisco

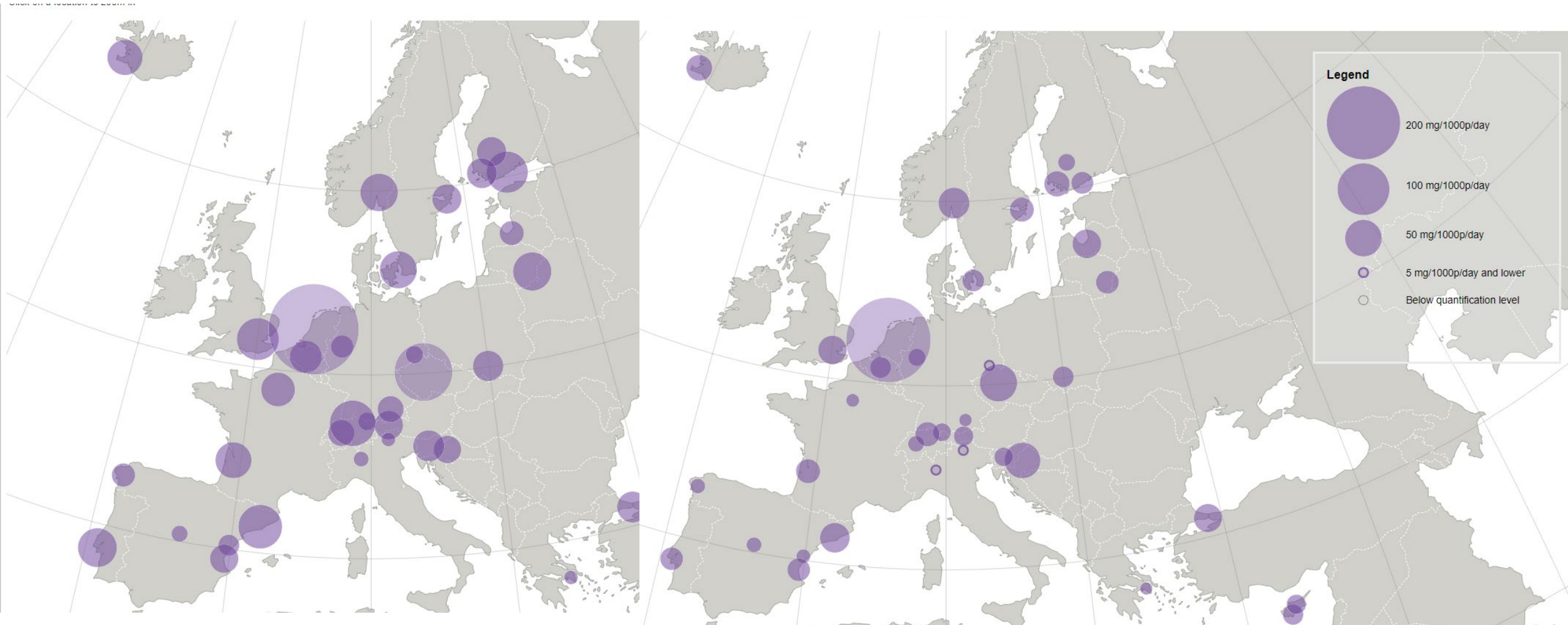


We can look at wastewater to understand human activity in a given service area

- We can detect usage of different consumer products
- Can detect consumption rates of certain foods
- Traces of pharmaceuticals can be found
- We can detect pathogens which tells us about infection rates
- Note, this is raw influent before treatment



We can look at wastewater to understand human activity in a given service area



MDMA in European sewer systems, weekends (left) vs. weekdays (right)

SARS-CoV-2 or Covid-19?

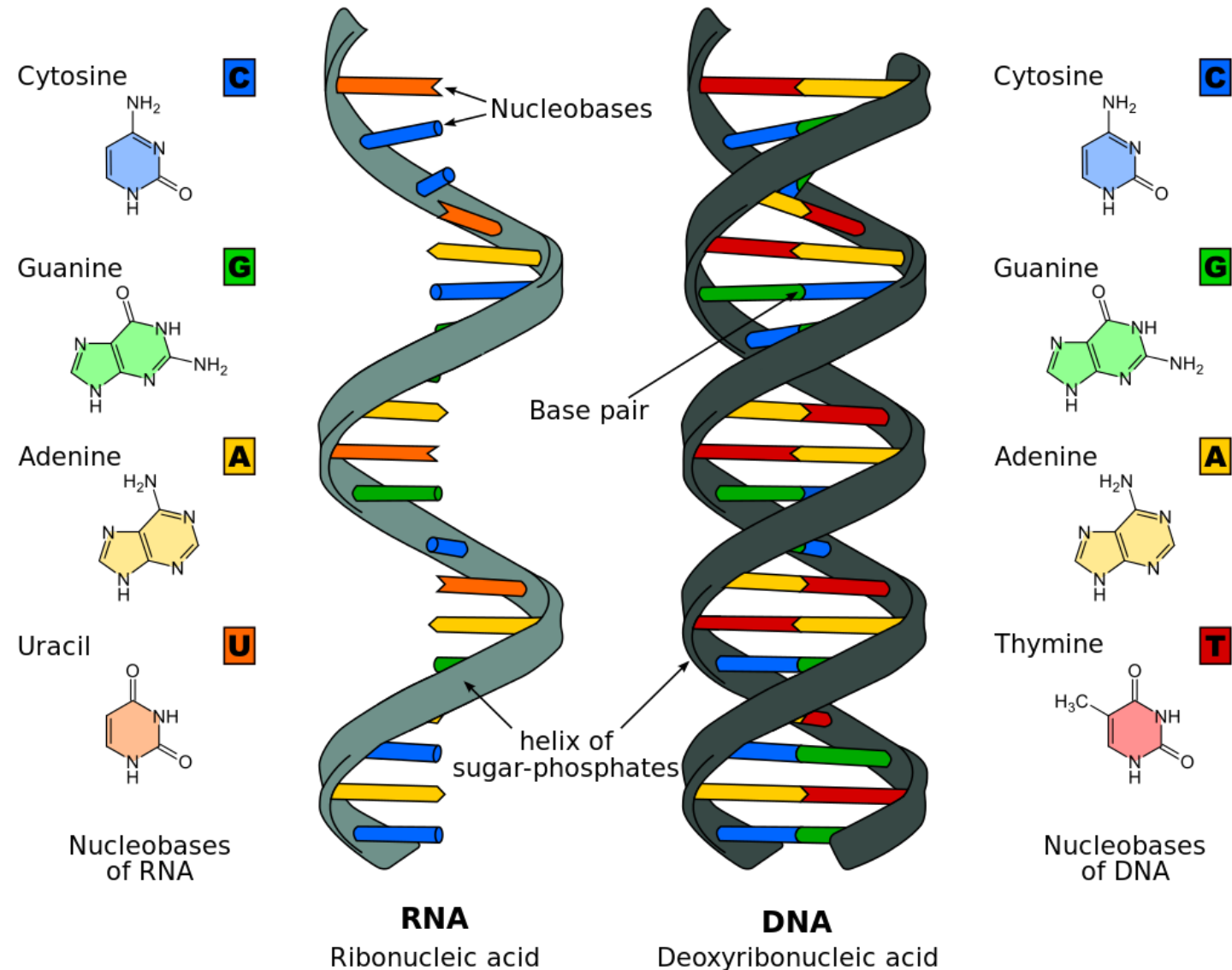
- SARS-CoV-2 is the name of the virus
- There are different types of coronavirus, SARS-CoV-2 is one type
- Covid-19 is the name of the disease

SARS-CoV-2 monitoring basics

- Infected individuals (symptomatic and asymptomatic) shed the virus in feces
- The virus dies off quickly in wastewater, but the RNA (genes) of the virus can still be detected using qPCR
- Water Environment Federation – “Researchers have yet to find evidence of survival of infective COVID-19 virus in feces and in wastewater systems”
- CDC, WHO, WEF – Wastewater is not a vector for transmission of SARS-CoV-2
- Raw wastewater is dangerous, but not because of SARS-CoV-2



Looking for genes



qPCR....quantitative Polymerase Chain Reaction

- Looking for a known part of the SARS-CoV-2 RNA
- A primer is introduced that locks onto that specific part of this genetic code
- Spare ingredients are introduced
- A polymerase is introduced which then copies the specific strand

VIRAL RNA: 5'-UGACCUGAAGACUGA-3'
3'-ACTGGACTTCTGACT-5'

VIRAL RNA: 5'-UGACCUGAAGACUGA-3'
3'-ACTGGACTTCTGACT-5'



qPCR

- Fluorescence is added which highlights the strands
- The step is repeated multiple times

VIRAL RNA: 5'-UGACCUGAAGACUGA-3'
3'-ACTGGACTTCTGACT-5'

VIRAL RNA: 5'-UGACCUGAAGACUGA-3'
3'-ACTGGACTTCTGACT-5'

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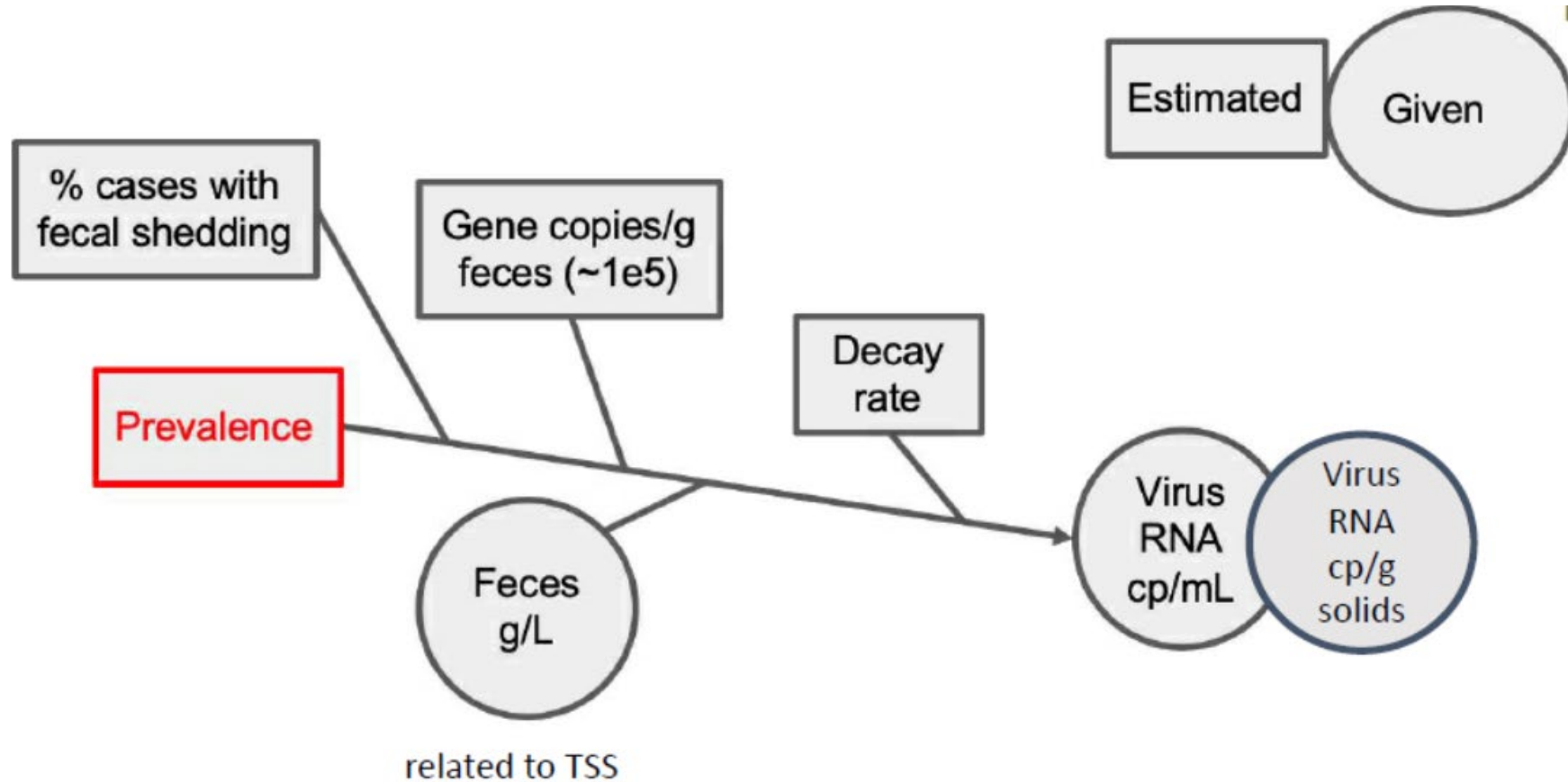


SARS-CoV-2 monitoring basics

- Not looking for the virus, looking for gene copies
- qPCR can tell us the concentration of gene copies per volume of wastewater
- Usually results are reported in gene copies/L or gene copies/mL

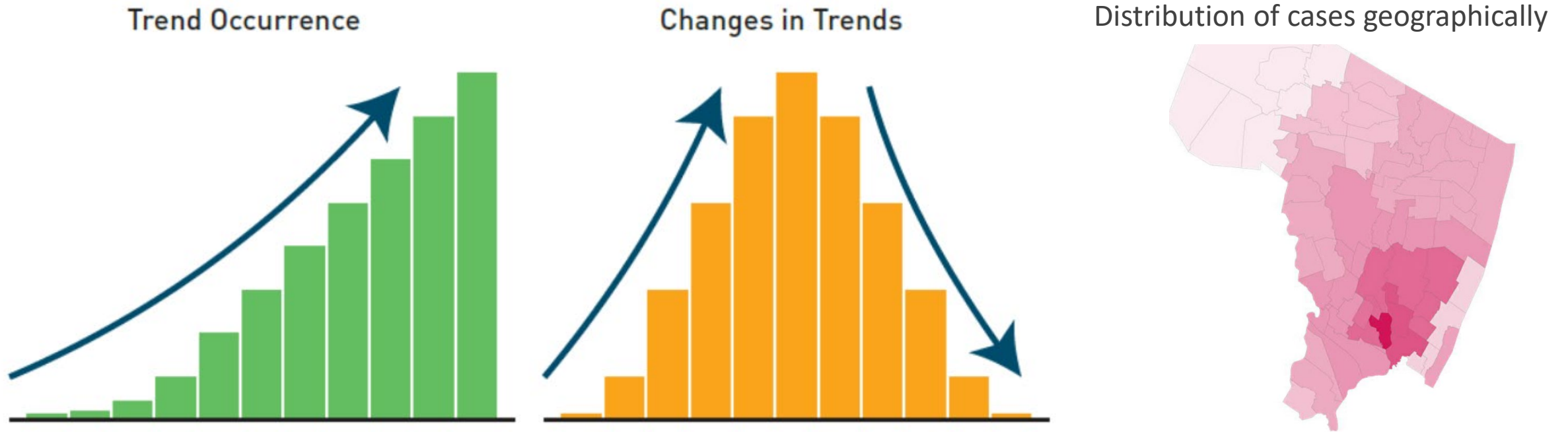


What wastewater surveillance can/cannot do



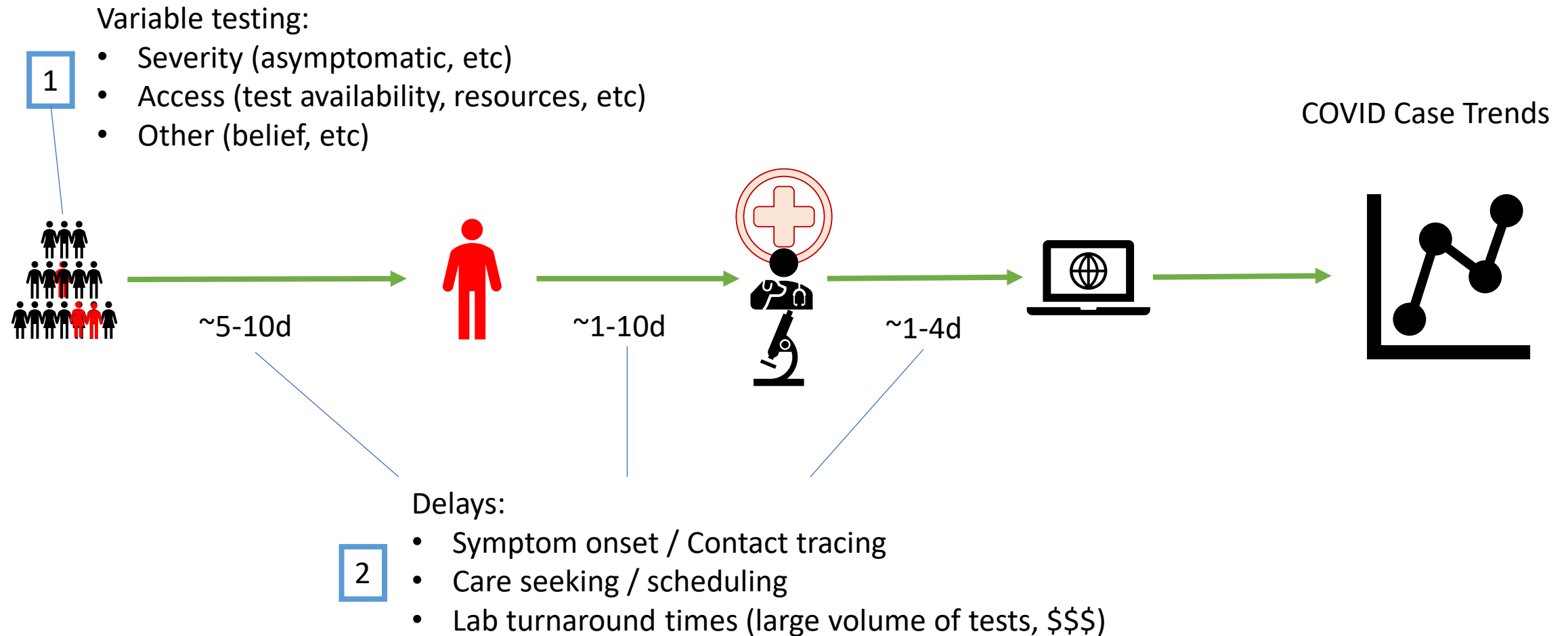
Currently, it is difficult for wastewater surveillance to calculate the number of infected individuals in a service area

What wastewater surveillance can/cannot do

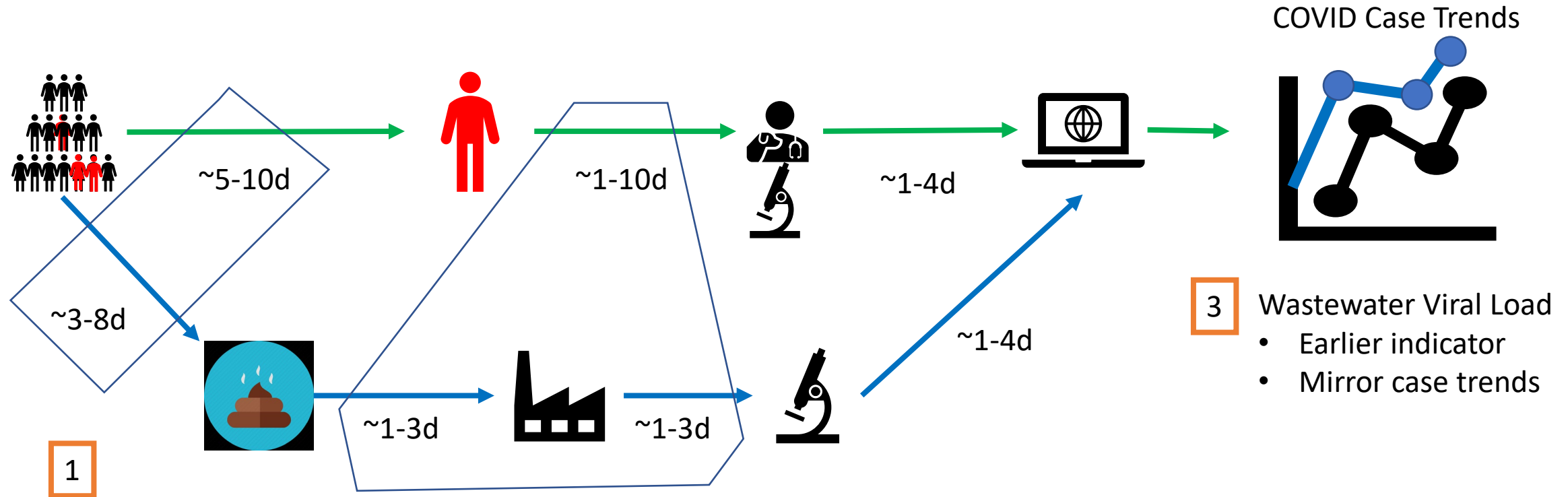


- Early warning of infections in locations that have none
- Early warning of changes in infection trends
- Areas where there are more infections
- Genetic ID of virus – where the strain came from

Case-Based COVID-19 Surveillance



Wastewater Based COVID-19 Surveillance



Everybody poops*

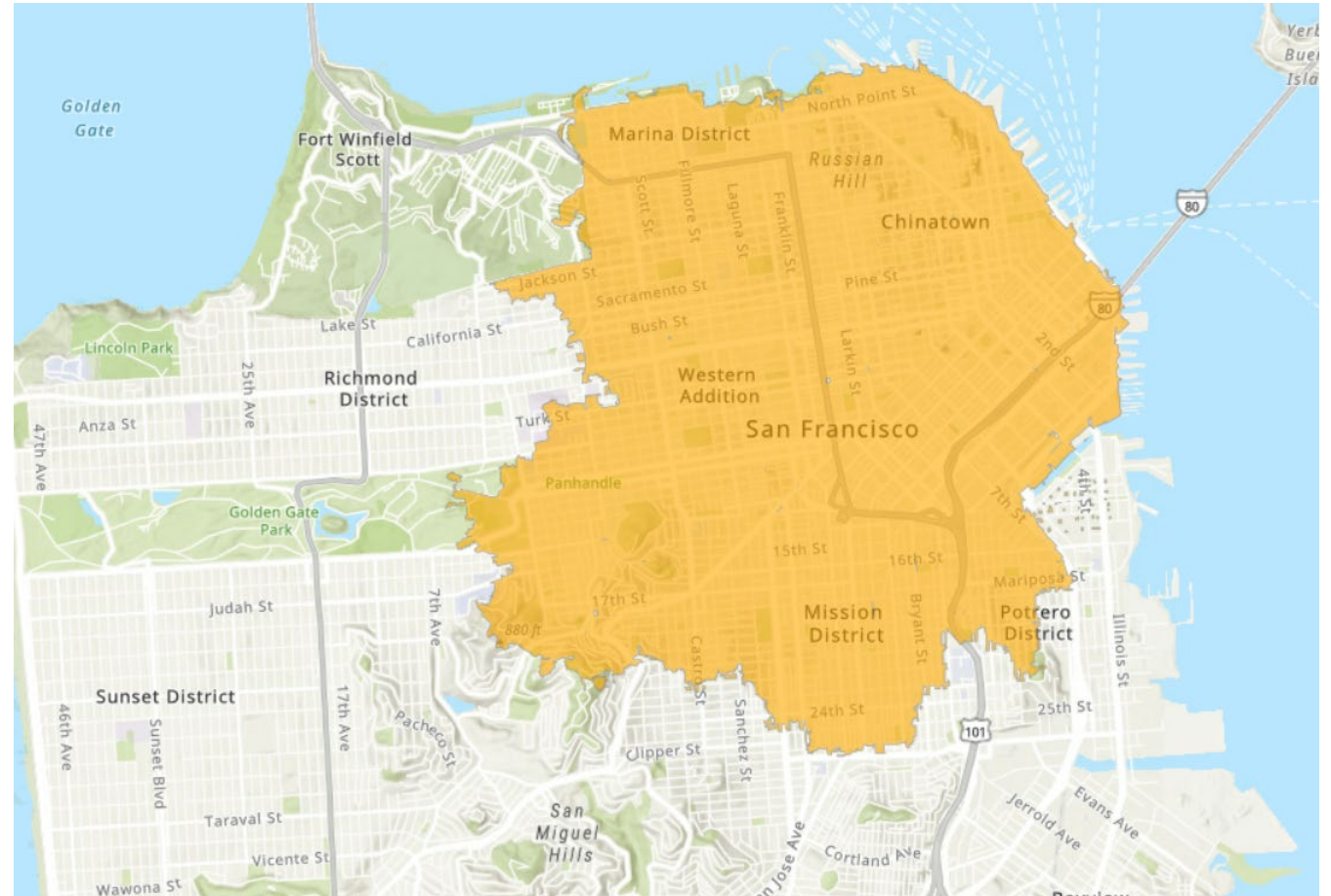
- *Not all poop goes to a WWTP
- *Not all infected will fecally shed

Less delay:

- 2
- Fecal shedding likely earlier than symptom onset
 - Time to reach WWTP < care seeking for tests

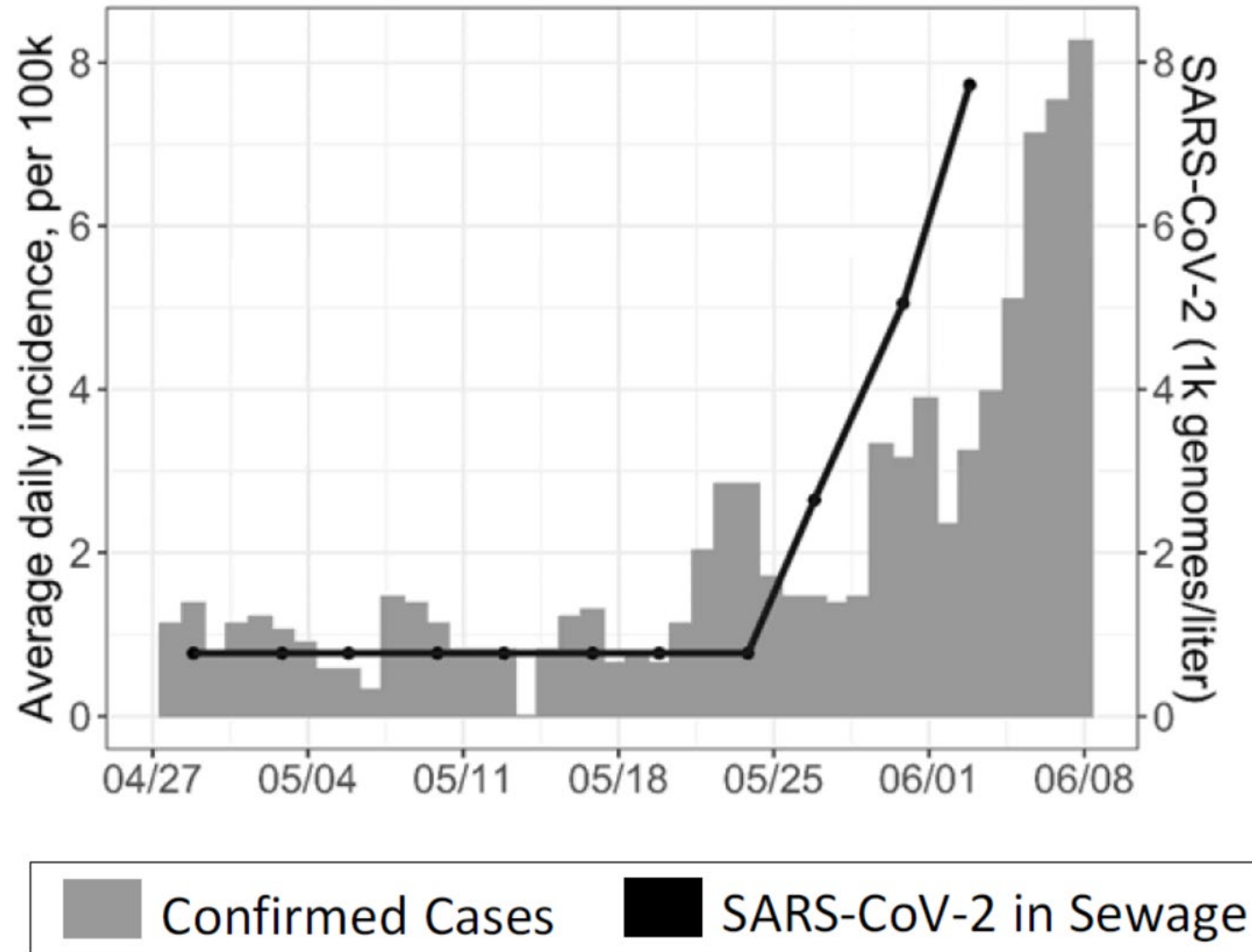
Wastewater Based COVID-19 Surveillance

- Testing an entire population would be costly and logistically impossible
- Wastewater sample for the service area will provide data for the entire population in the service area
- One test vs. 200,000



How can this tool be used?

- Early warning
- Wastewater surveillance predicted the Memorial Day surge in the Southwest
- Monitoring for early warning requires more frequent monitoring



How can this tool be used?

- Screening for presence of the virus, especially in a residential facility
- Positive signal found in a dorm, all residents tested, two positive cases found and isolated

The Washington Post

Democracy Dies in Darkness

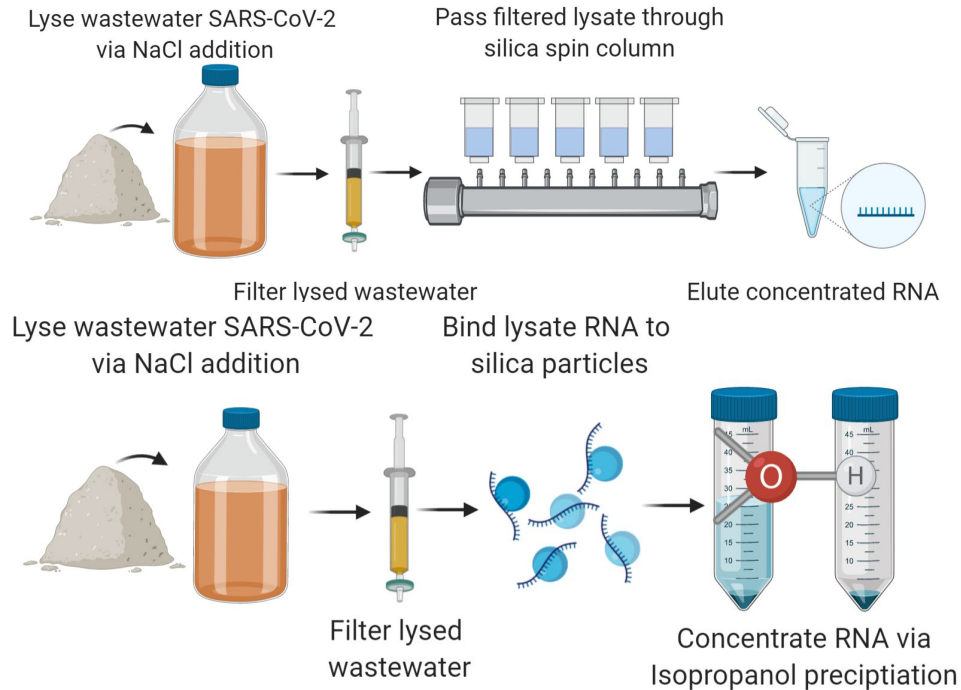
The University of Arizona says it caught a dorm's covid-19 outbreak before it started. Its secret weapon: Poop.

SFPUC partnered with University Researchers

- Sending samples to UC Berkeley and Stanford weekly since July and March respectively
- Researchers needed to first finalize methods for quantification of gene copies/mL



UC Berkeley



Direct wastewater RNA capture and purification via the "Sewage, Salt, Silica and SARS-CoV-2 (4S)" method V.1

Direct wastewater RNA extraction via the "Milk of Silica (MoS)" method - A companion method to "Sewage, Salt, Silica and SARS-CoV-2 (4S)"

- UC Berkeley Lab methods available on protocols.io (google titles above)
- Methods avoid using RNA extraction kits which are in short supply

How to determine strength of waste

- Infiltration and Inflow
- Non sanitary flow
- Stormwater in a combined system
- PMMoV used as a marker of sanitary flow
- Pepper mild mottle virus – by far the most common RNA virus in poop of healthy individuals
- In a study of 12 treatment plants, concentration in raw wastewater was over 1 million copies per *milliliter* at every plant



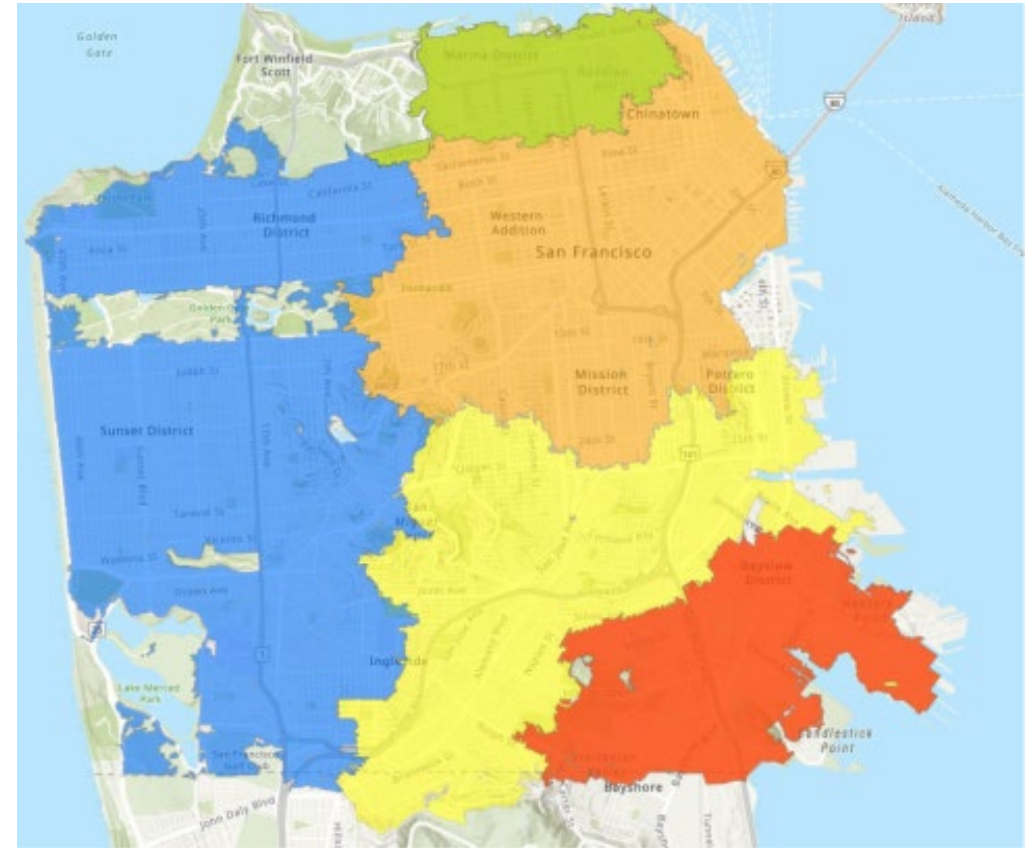
SFPUC's role

- SFPUC does not manage results from testing
- SFPUC is the wastewater expert
- SFPUC provides sampling and flow expertise



Sampling locations

- Can divide city into 5 subregions
- Sampling at two treatment plants
- Sampling at three pump stations
- Sampling at one major trunk line



Sampling locations

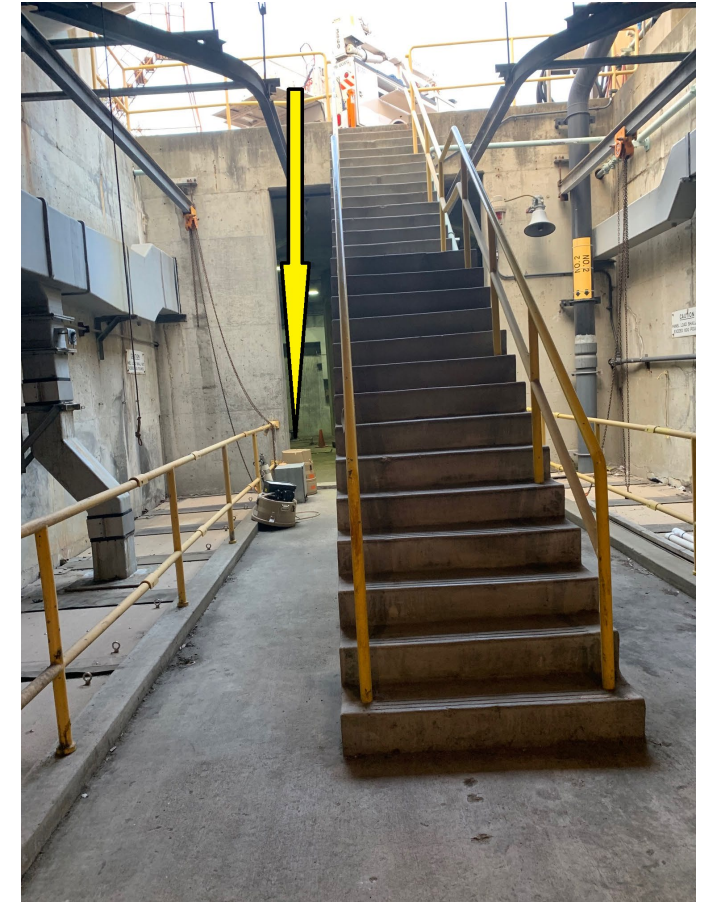
- Pilot sampling project at a skilled nursing facility
- This is coupled with monthly testing of residents and staff
- Goal is to provide an early indication of an infected individual
- Risk of false negative, less concern about false positive

Sampling logistics

Residential facility

vs. trunk line

vs. pump station



Sampling logistics

- Leveraged existing field monitoring expertise and equipment for sampling set up



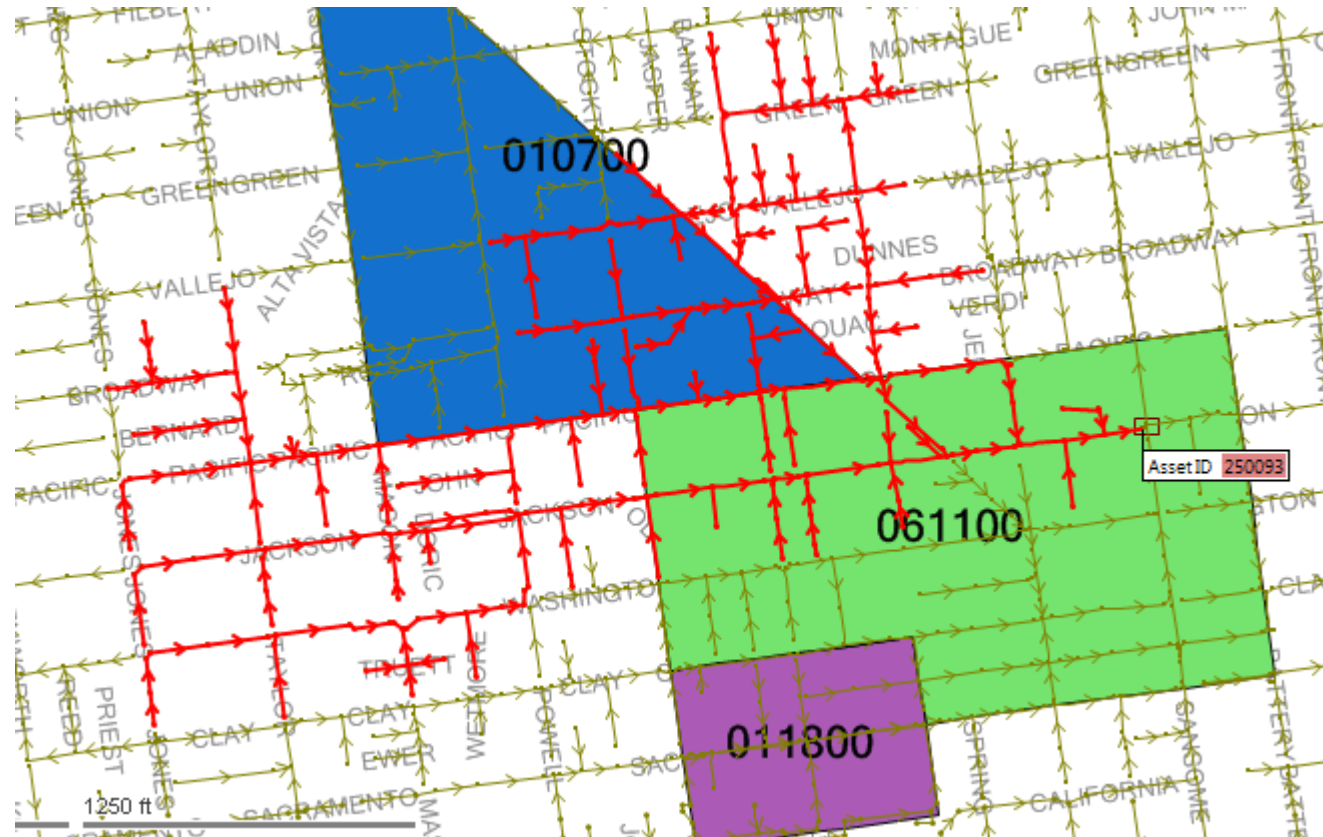
Sampling logistics

- Field work often presents unforeseen challenges



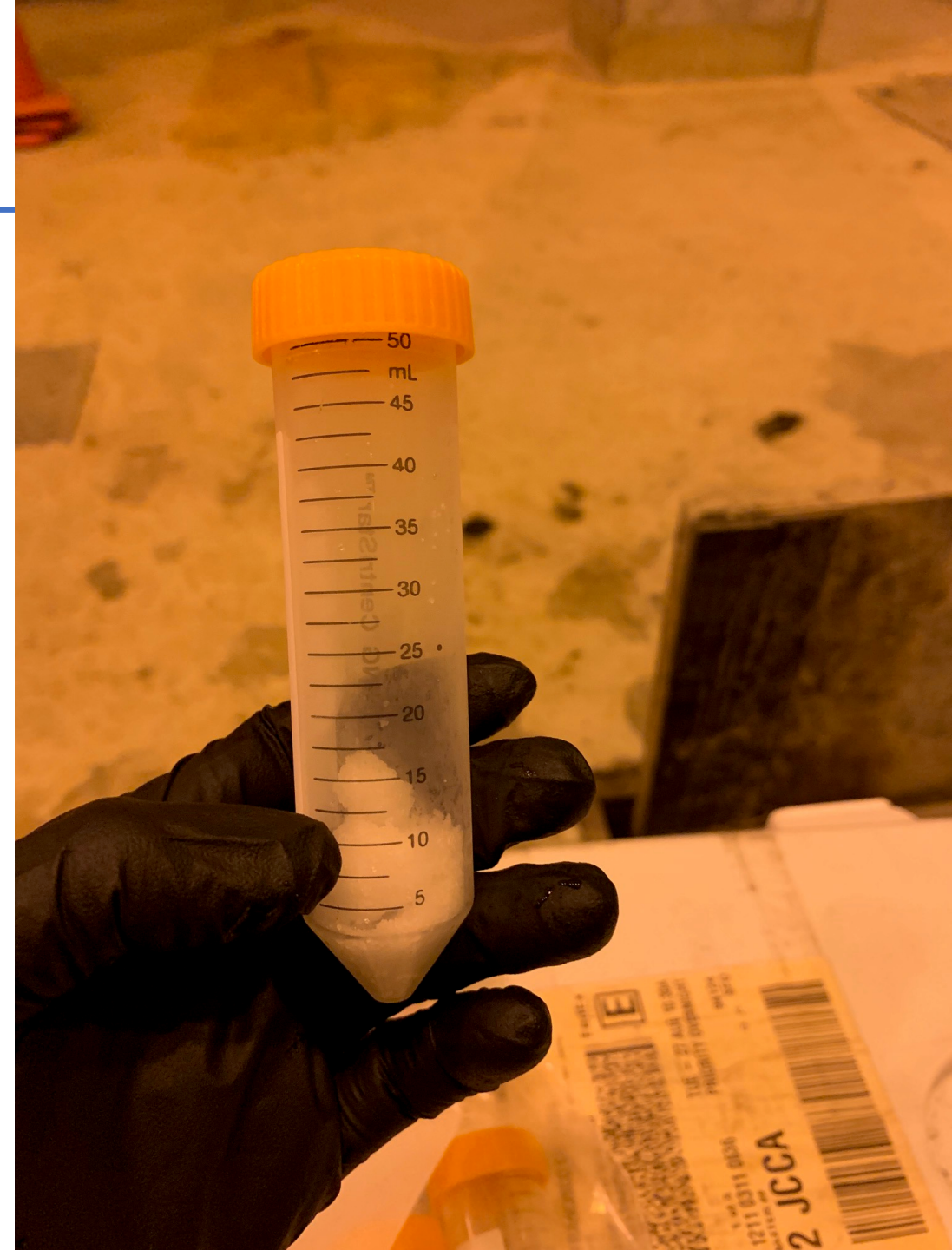
Sampling logistics

- Not every location or service area can be neatly sampled



Sampling logistics

- All samples are 24 hour composite
- Sampling plant influent is from daily composite collected by lab
- Field monitoring requires setting up ISCO sampler 1 day prior with ice
- 10 L of sample is mixed and poured into four sample tubes



Covid-WEB

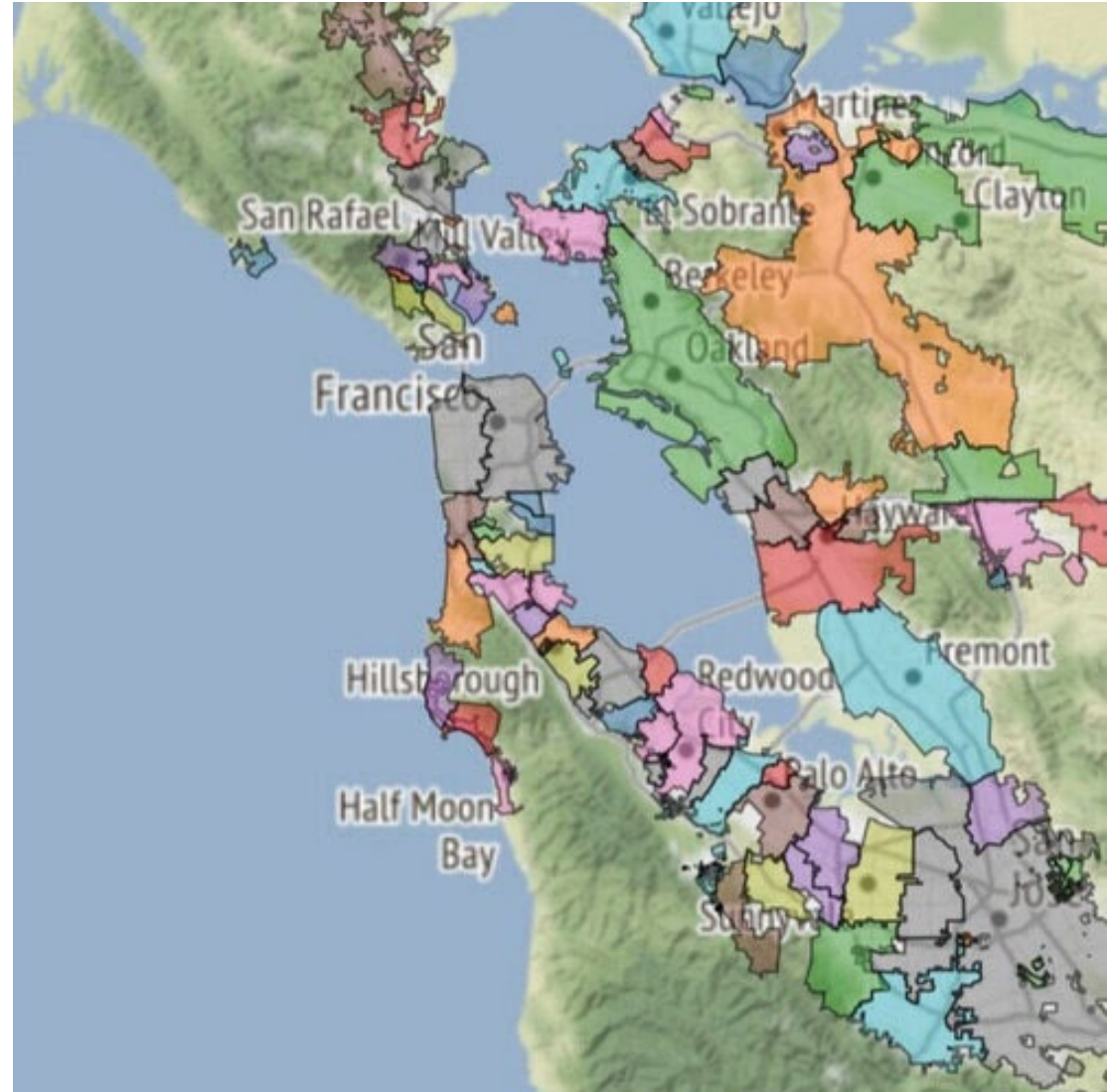
- New lab can now sample up to 25 samples/week from San Francisco
- Capacity to run 100 samples/day from Bay Area
- UC Berkeley spearheading Covid-WEB (Wastewater Epidemiology for the Bay Area)
- www.covid-web.org/



Covid-WEB

Agencies from the following counties currently sending samples

- San Francisco
- Marin
- Santa Clara
- Contra Costa
- Alameda



Sampling costs

- In kind costs for sampling collection
- UC Berkeley has funding for this work
- Lab analysis covered by UC Berkeley funding
- Looking for funding to add staffing resources

Key Takeaways

- Covid-19 does not spread through wastewater
- Efforts to culture live, infectious SARS-CoV-2 in wastewater have not been successful
- Fragments of SARS-Cov-2 genes can be found and measured in wastewater
- Wastewater surveillance is another tool for monitoring the spread of Covid-19

Thank you – questions?

