Wastewater Monitoring for Public Health

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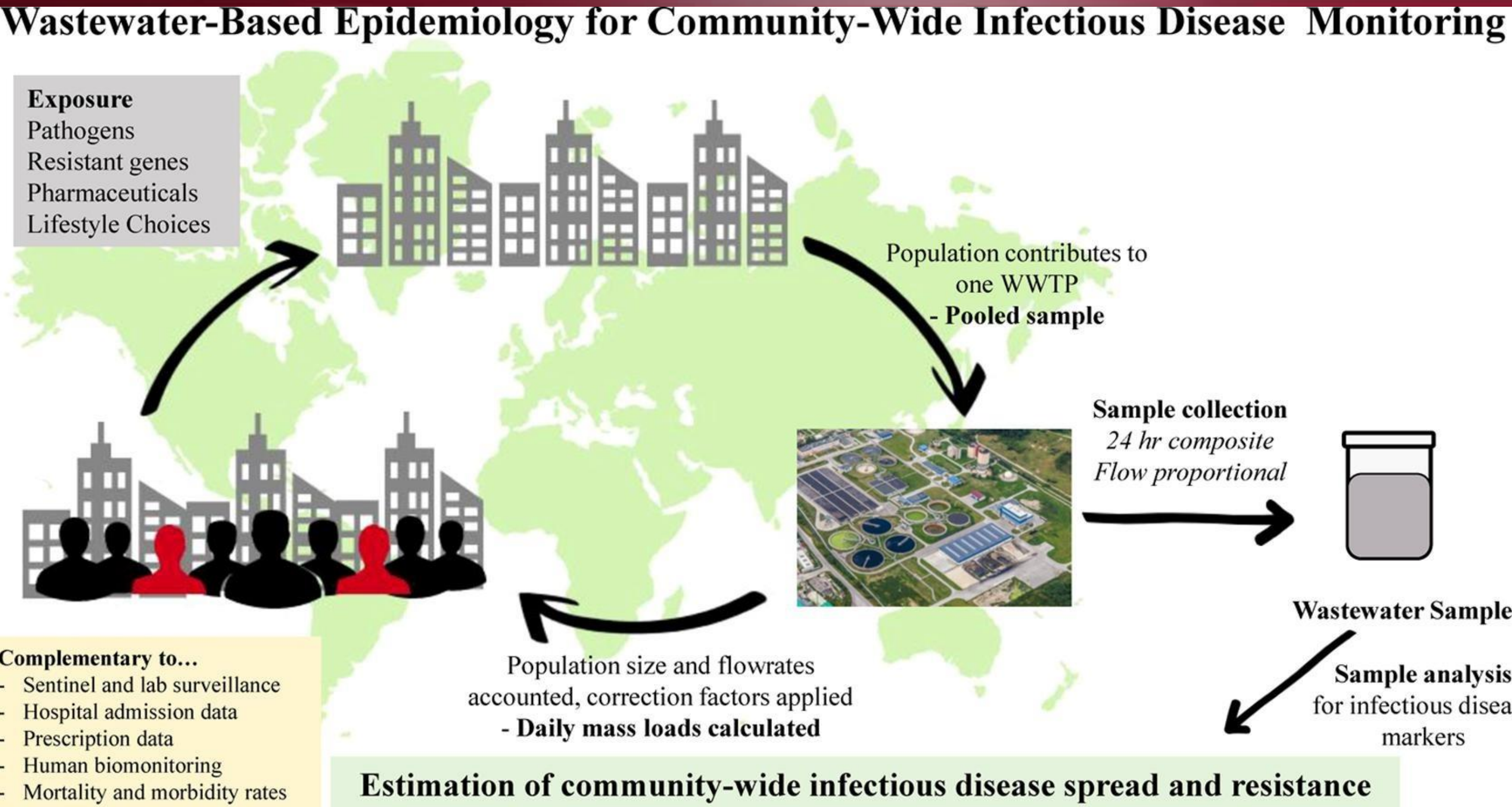
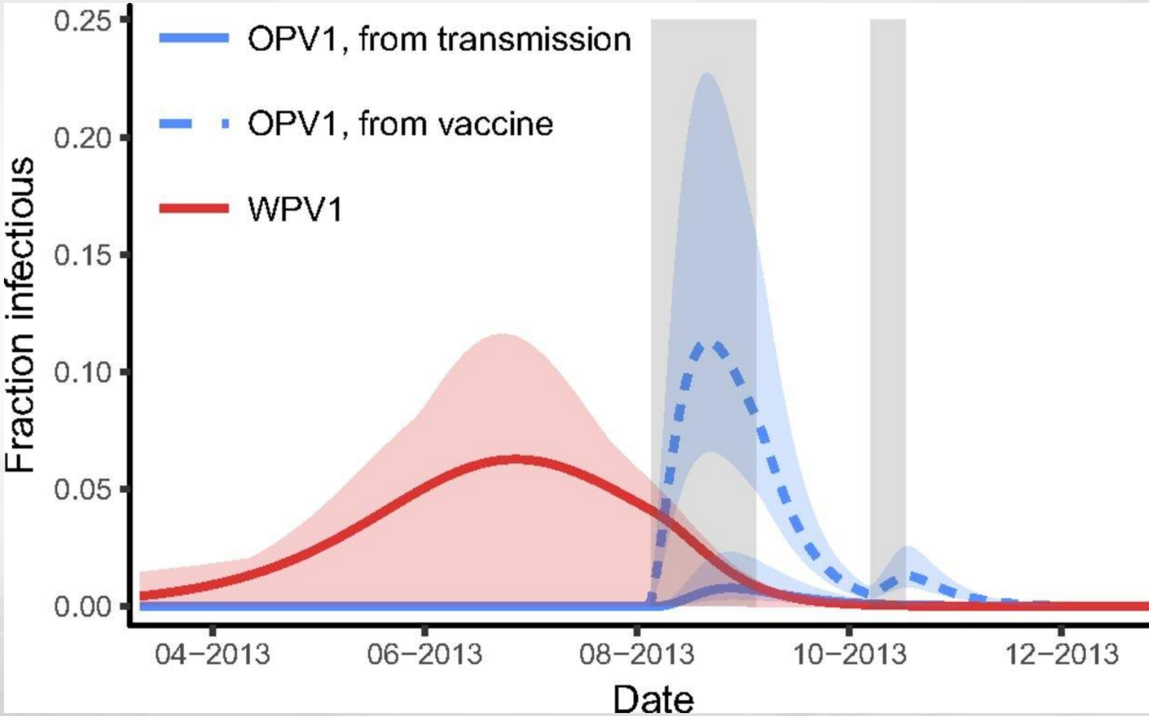
OTCO Annual Class III & IV Workshop for Water and Wastewater Operators
8/11/2022



Natalie Sims, Barbara Kasprzyk-Hordern,2020, Future perspectives of wastewater-based epidemiology: Monitoring infectious disease spread and resistance to the community level, *Environment International.*

History of wastewater/environmental monitoring for public health

- Israeli health department set up in 1989 sewage monitoring to detect poliovirus
- Tracking poliovirus transmission and prevent outbreaks of polio-caused paralysis (WHO, 2003)



Brouwer, Andrew F., et al. "Epidemiology of the silent polio outbreak in Rahat, Israel, based on modeling of environmental surveillance data." *Proceedings of the National Academy of Sciences* 115.45 (2018): E10625-E10633.

Wastewater monitoring early on during COVID-19 pandemic

- SARS-CoV-2 measured as RNA gene fragment (most likely inactive)
- SARS-CoV-2 RNA viral gene fragments get to wastewater mainly from stool of infected people

Crank et al., 2022, STOTEN, 806: 150376

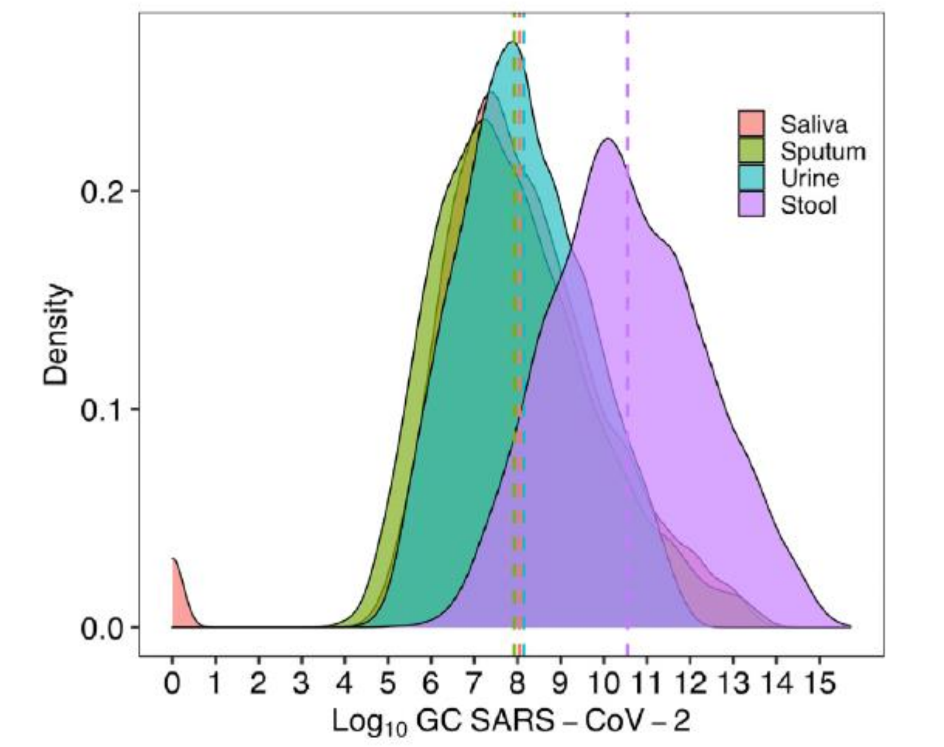
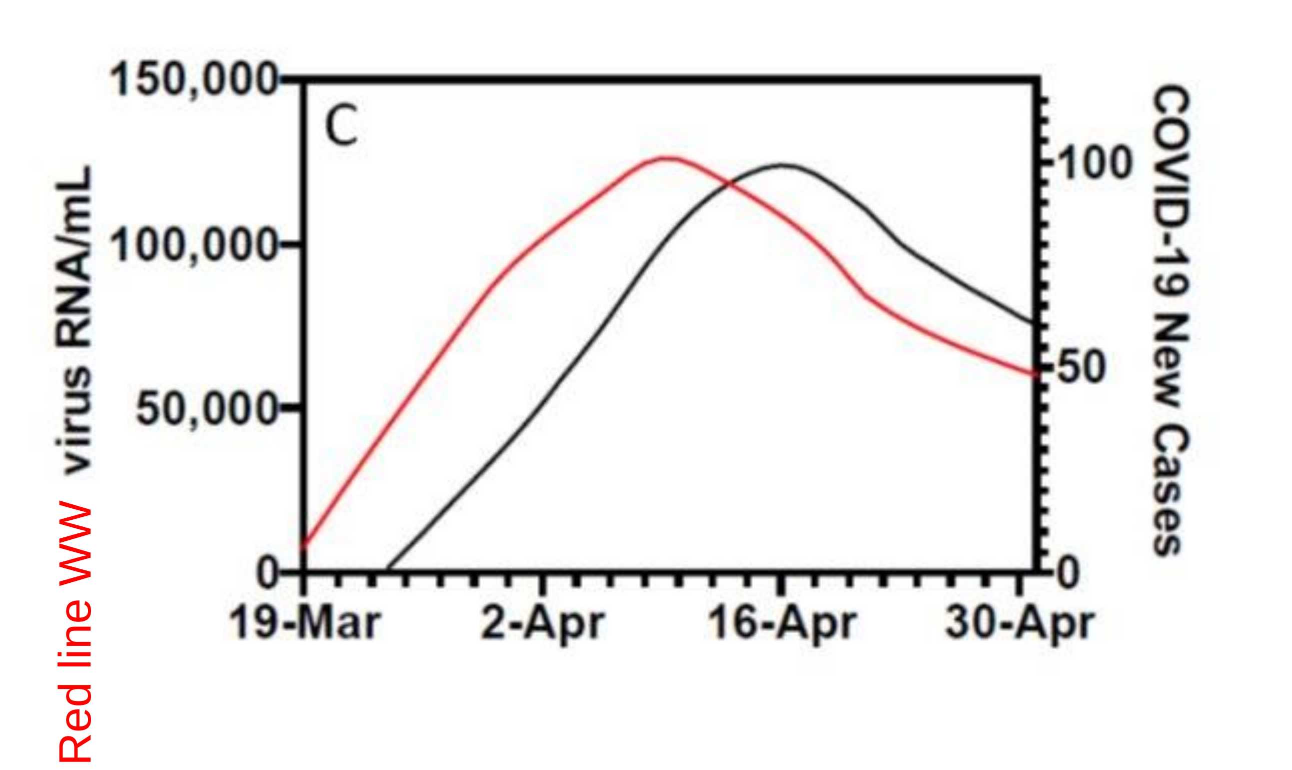


Fig. 1. Daily viral load of SARS-CoV-2 produced to a sewer system by a population of 1000 SARS-CoV-2 infected individuals stratified by shedding route. The dotted vertical line represents the mean.

Wastewater monitoring early on during COVID-19 pandemic

- Wastewater concentration is correlated with new cases
- Wastewater is used as a leading indicator



Peccia et al, 2020, https://doi.org/10.1101/2020.05.19.20105999

Ohio's Wastewater Monitoring Timeline

May 2020
Initiation of Network
Governor DeWine initiates wastewater SARS-CoV-2 monitoring project

Funding of Network
June 2020
Ohio EPA – uses CARES funds for WW monitoring
ODH is project lead

July 2020
First Sites Onboarded
7 large cities
15 locations sampled
3 laboratories

Onboarding Additional Sites
August-October 2020
Recruit medium and smaller cities
4 added laboratories
46 sites added

November-December 2020
Technical Aspects
Adding sites
Build spatial join with sewershed cases
Dashboard graphs and file downloads

Expand the Network
January-May 2021
CDC funding
Statistical data analytics
Build sequencing framework

May–current 2021-2022
Improve the Network
ODH only laboratory
Statewide sample delivery
Advancement of analytical tools

Ohio Department of Health

Partners during Network Initiation

Ohio Department of Health

Ohio Environmental Protection Agency

Water Resources Center Ohio WRC

United States Environmental Protection Agency

The University of Toledo

BGSU

Best Courier

The University of Akron

Kent State University

Battelle

Ohio Section American Water Works Association

American Expediting

CDC Centers for Disease Control and Prevention

Mercury Services

OWEA Ohio Water Environment Association

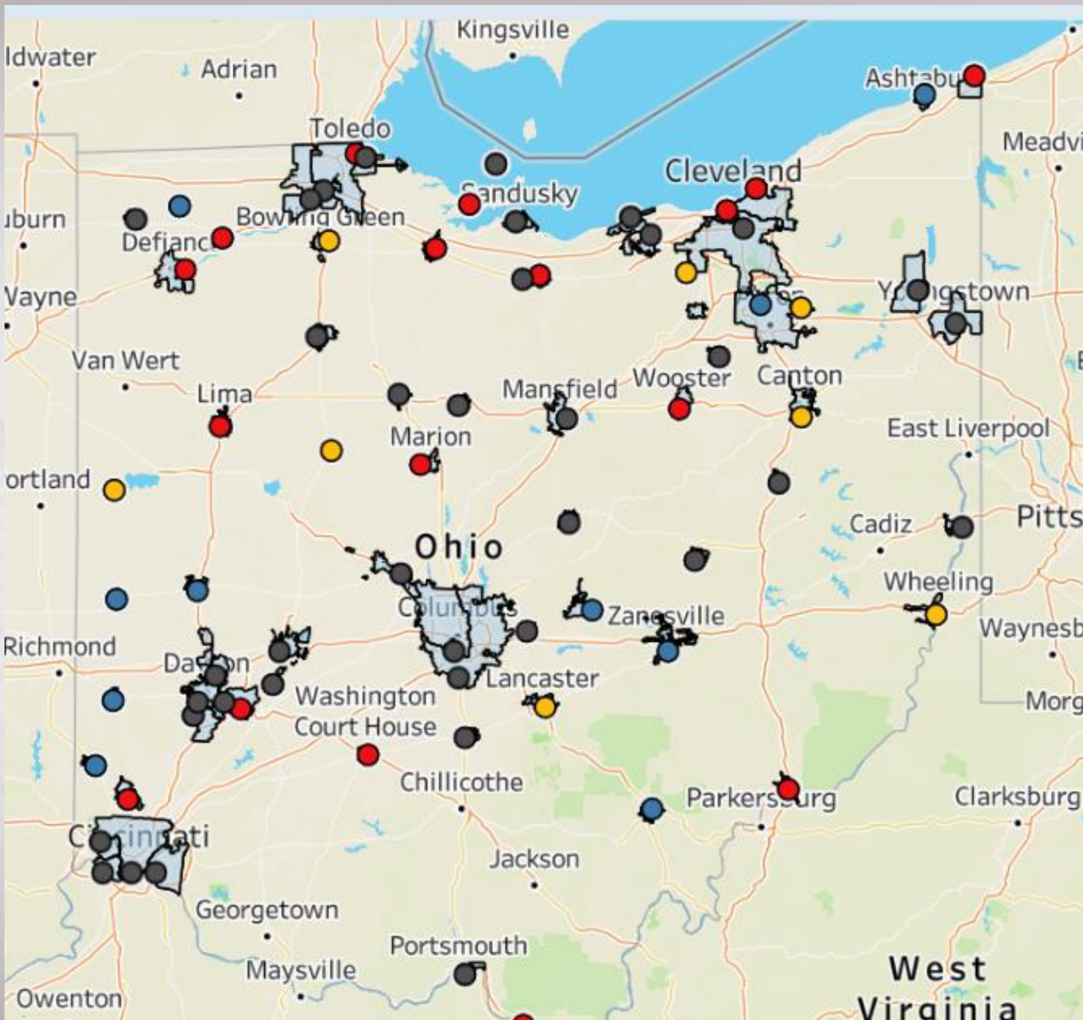
Environmental Training Center OTCO Since 1964

LuminUltra microbial monitoring

Microbac

The Ohio State University

Ohio's Wastewater Monitoring Locations



- Twice weekly monitoring of 70+ locations
- Monitoring covers approximately 5 million of Ohio's residents
- Monitoring covers residents in 54 of Ohio's counties
- The populations served per treatment plant range from 3,300 – 655,000.
- Wastewater treatment plant flows range from 0.6 – 130 MGD

Ohio Department of Health

Weekly Schedule

Sampling

- Utility collects influent sample twice a week, supplies are provided
- Sample stored in fridge temperature until sample pick up (few hours)
- Sample picked up by courier, supplies dropped off for next sample

Analysis

- July/2022 only ODH Public Health Lab
- One consistent method
- Lab analyses quantification in 24-48 hours

Application

- Data from utility and lab compiled
- ODH dashboard update
- Trend analyses and alerting communities

Wastewater Collection System

- Flow of wastewater begins at homes and businesses
  - Water moves to treatment plant via gravity and pumping stations
- Residence time in sewers varies based on distance of origin
  - Closer to plant = short residence time
  - Further from plant = longer residence time
- Long residence time, wastewater pH, and wastewater temperature cause RNA molecules to decay

Case diagnosis date

Case location in relation to sampling spot

	-2 to 6 days	-2 to 13 days
0 to 4km	0.76	0.67
0 to 9km	0.71	0.60
0 to 14km	0.70	0.60
0 to 19km	0.64	0.56
0 to 24km	0.64	0.57
0 to 29km	0.64	0.57
0 to 34km	0.64	0.57

The numbers and blue color represent the probability of the case detection in wastewater based on distance in collection system and disease onset time

Black J. et al., Epidemiological evaluation of sewage surveillance as a tool to detect the presence of COVID-19 cases in a low case load setting. Sci Total Environ. 2021. doi: 10.1016/j.scitotenv.2021.

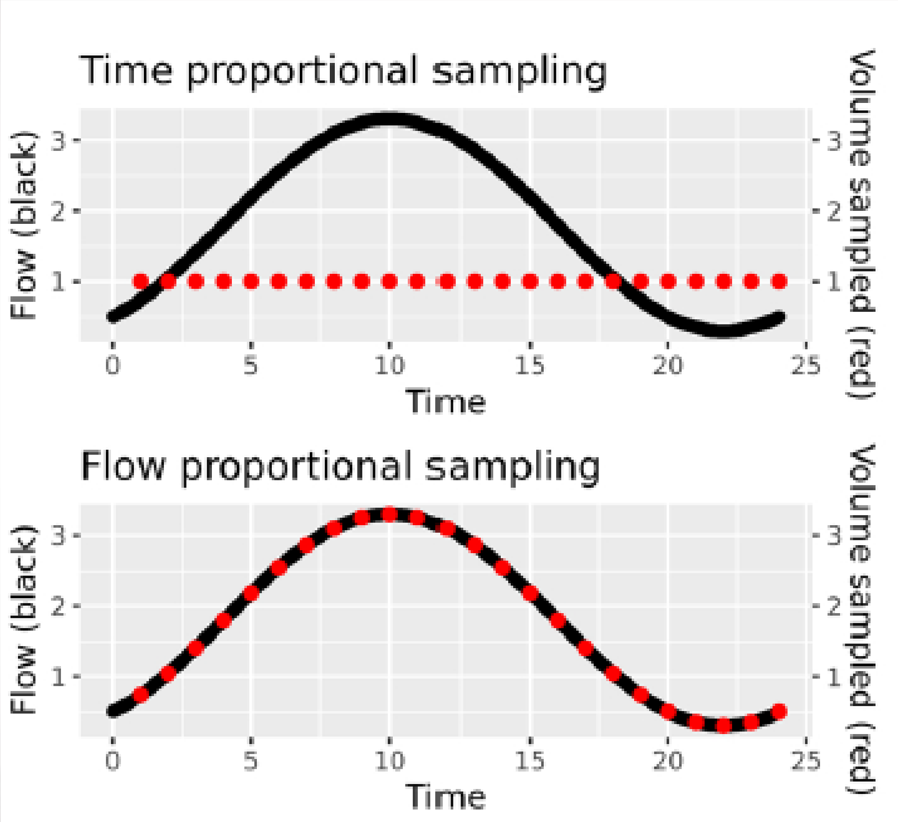
Influent Sampling Configuration

- Types of samples
  - Single Grab
  - Multiple Grab
  - Composite
    - Flow weighted
    - Time weighted

Photos courtesy of Terry Korzan from Elyria WWTP and Jim Boyd from Beavercreek WWTP

Composite sampling

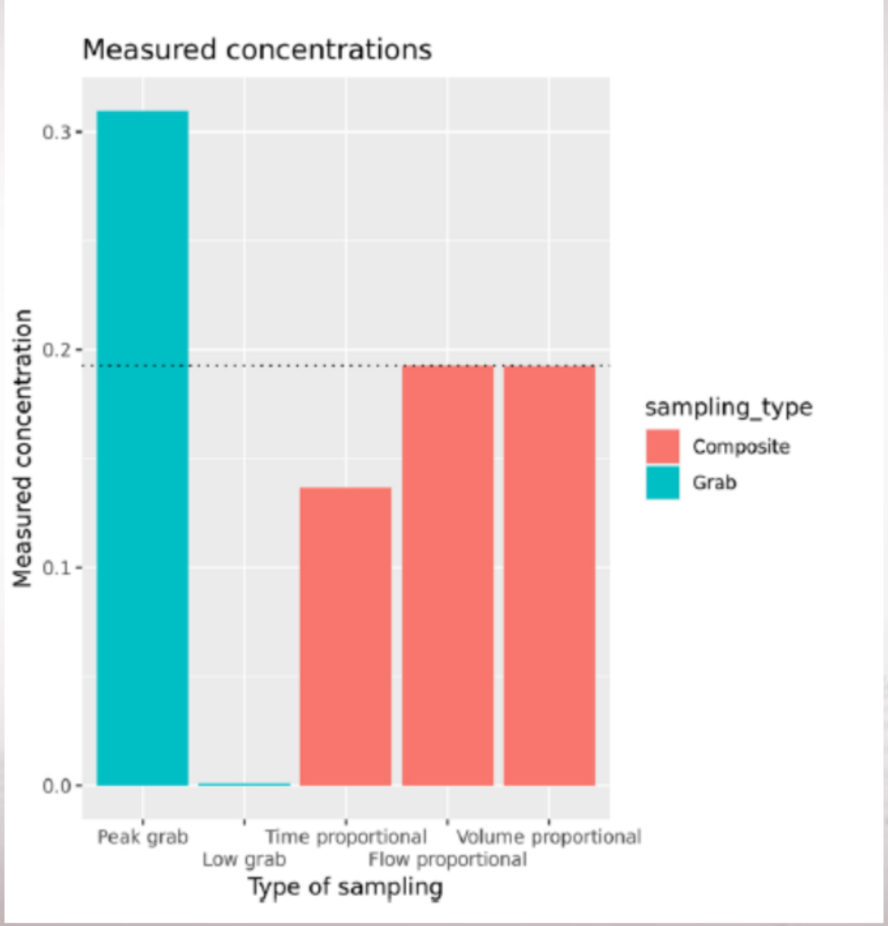
- Time Proportional
  - Even time spacing of samples
  - E.g. 1 Sample @ 15 min intervals
    - 96 samples
- Flow proportional
  - Spacing of samples based on amount of flow
  - Calculate by taking Avg. Daily Flow divided by 96 samples
    - 7.5 MGD / 96 samples
    - @ 78,000 gallons between samples



Wade, Matthew J., et al. "Understanding and managing uncertainty and variability for wastewater monitoring beyond the pandemic: Lessons learned from the United Kingdom national COVID-19 surveillance programmes." *Journal of Hazardous materials* 424 (2022).

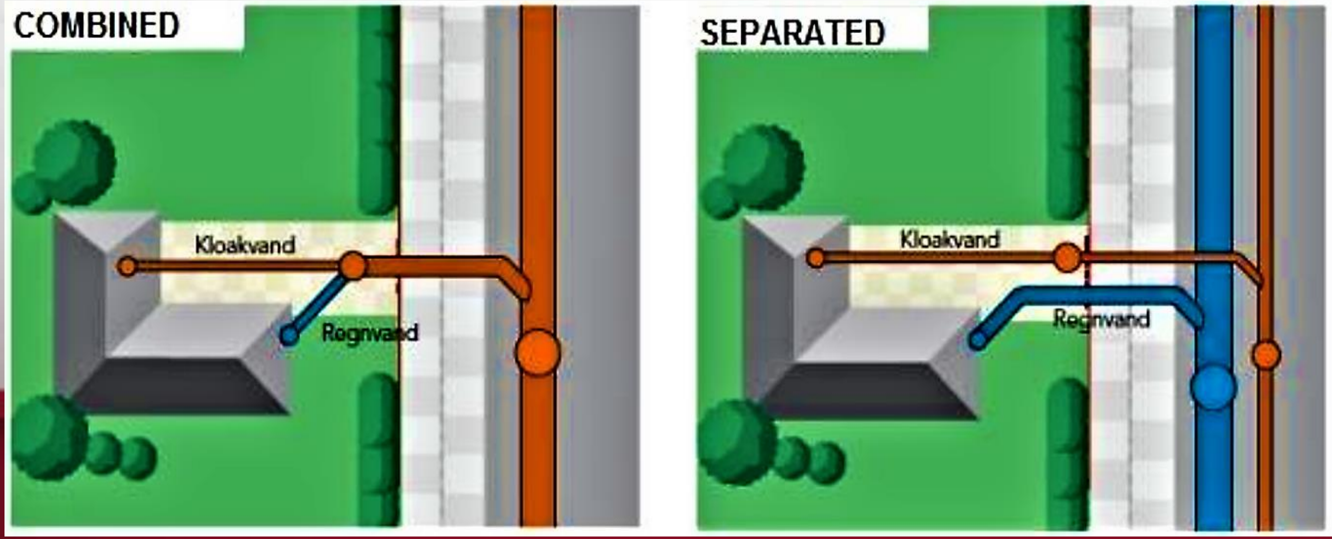
Effects of Sampling Types

- Grab sample
  - Artificially high or low numbers depending on flow
- Time Proportional
  - Better representation of flow than Grab samples, but still could artificially affect numbers
- Flow Proportional
  - Best representation of flow, as it reflects actual conditions and concentrations



Stormwater and Concentration Dilution

- Combined Sewers (sewer + stormwater)
  - Stormwater flow could vary based on event
  - Allow for dilution of the parameter being tested
- Closed Sewers (sewer only)
  - Designed to transport only sewage
  - Not a perfect system (Inflow & Infiltration is typical)



This picture shows the difference between a combined and a closed (separated) sewer system. After the combined system is replaced, sewage and stormwater can be managed in two separate systems. Source: LADEN (2010)

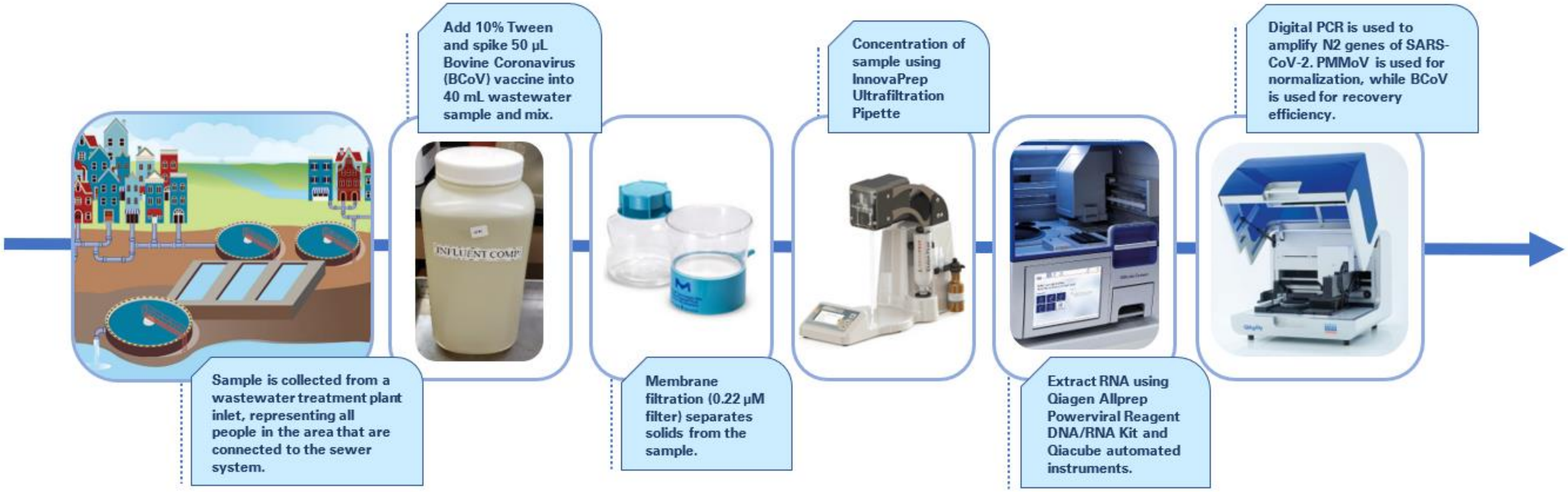
Samples and Supporting Data Collected

Crucial information used to normalize the results for possible dilution due to rain

Parameters that help evaluate possible RNA degradation

Sample Collection Information:	
Site ID (Ohio EPA NPDES Permit number):	
Facility Name:	
Sample Collection Type (select)	☐ Grab ☐ Time-weighted composite ☐ Flow-weighted composite
Sample Start Date (day that sample collection began, MMDDYYYY):	
Sample Start Time (time that sample collection began, in military/24-hour time):	
Sample Collection Duration (n/a for grab):	
Flow Volume (for composite) or Flowrate (for grab):	
24-hour Rainfall (inches or yes/no):	
OPTIONAL: Water Quality Measures of Influent Wastewater	
pH of influent wastewater:	
Temperature (C) of influent wastewater (min and max):	
Other(s):	
Additional Notes:	
Contact Information:	

Quantifying SARS-CoV-2 virus in wastewater

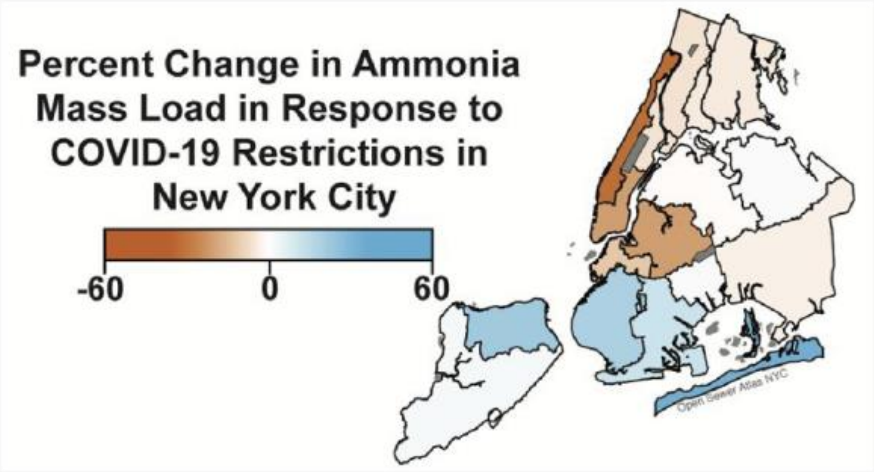


Method used by the ODH Public Health Laboratory

Normalization of the Sample Result

Aims to improve the estimate of the population contributing to wastewater

- Variability of flow within a sewershed
  - Holidays, weekdays vs weekends, storms
- Markers used for normalization
  - Population of sewershed
  - Flow rate of wastewater
  - Human fecal viral indicators
    - Pepper mild mottle virus [PMMoV]
    - Cross-Assembly Phage [CrAssphage])



Hoar et al., CS EST Water 2022, Publication Date: April 20, 2022
https://doi.org/10.1021/acsestwater.2c00052

Recovery efficiency

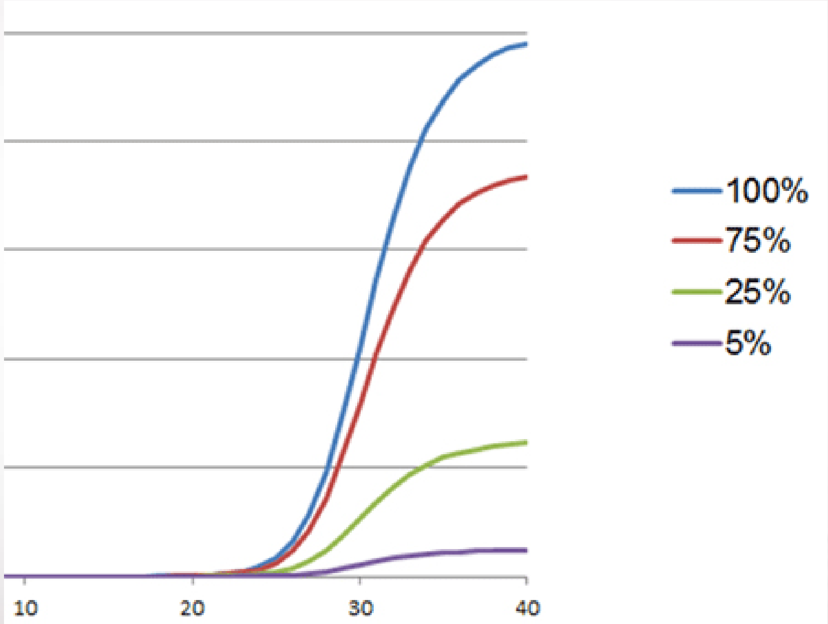
Low recovery may lead to underestimation of actual concentration

- Method used to concentrate and extract RNA may lead to RNA loss.
- Lab spikes viral surrogate to each wastewater sample and then evaluates the resulting concentration
  - Surrogate resembles target contaminant (BCoV, OC43)
  - Concentration of spiked surrogate is known
  - Surrogate must be something not present in wastewater
- Recovery efficiencies vary (1 – 60%)

PCR Inhibition

→ Leads to underestimation of the actual concentration

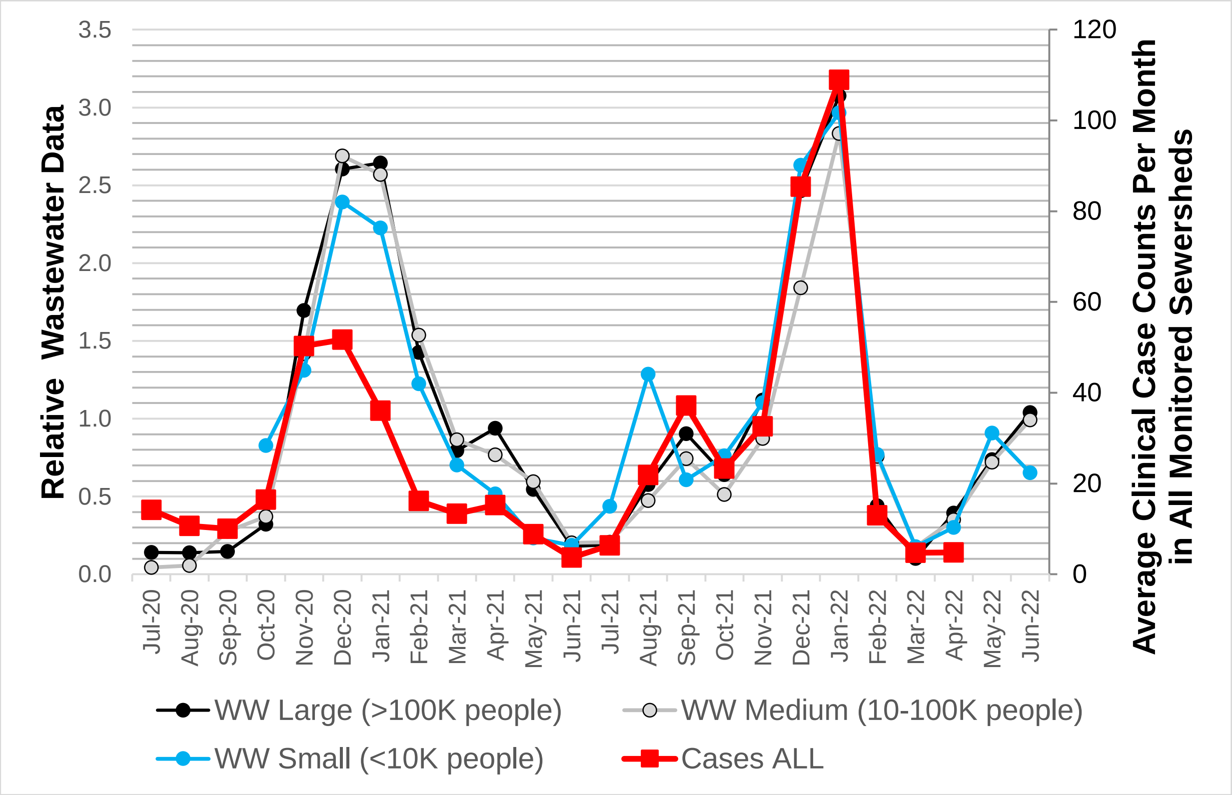
- Wastewater can contain known PCR inhibitors
- PCR inhibitors prevent amplification
- Dilution of sample could decrease inhibition
- Some available kits designed by companies include steps to remove inhibitors



Promega; https://www.promegaconnections.com/dealing-with-pcr-inhibitors/

Monitoring Results

- Cases are significantly correlated with wastewater concentrations throughout the network
- Wastewater concentrations lead time is site specific, but approximately 3 days on average



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Site Action Threshold – short term

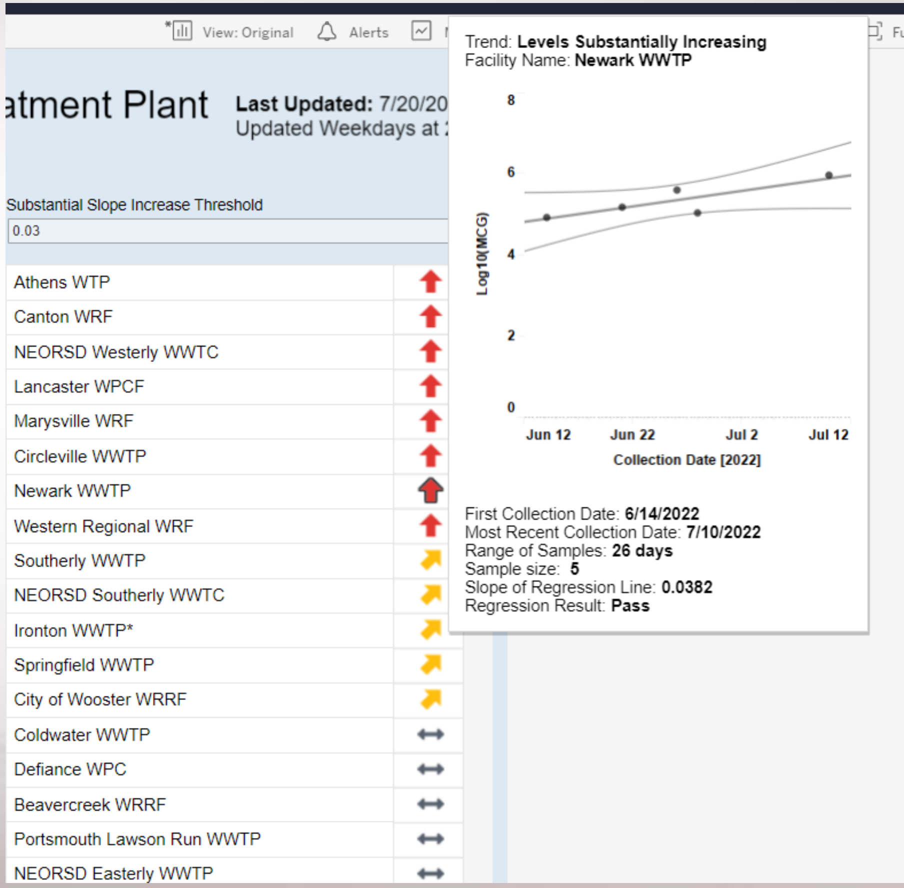
- Alert sent to local health district and wastewater utility when 3 samples show a sustained increase of at least 10-fold (1 log)
  - Can indicate increases due to a community event, such as a festival, concert, or college move-in
  - Can indicate the beginning of community spread

Fitzgerald et al. (2021) indicates:

- 25 cases detected in catchments >100 K
- 5 cases detected in catchments 10 – 100 K
- 1 case detected in catchment of <10 K

Site Action Threshold – long term

- 5-point regression (about 2.5 weeks) with a slope threshold of 0.03
  - Using log10 of million gene copy per day concentrations
- If the five samples meet this threshold -> indicates that community spread is likely
- Applies only for sites with twice weekly monitoring



Regional and Statewide Action Thresholds

- Currently under development
- Based on aggregating results from multiple facilities
  - Ohio Preparedness/Hospital regions
- Utilizing percentiles
- Used alongside other indicators (e.g., case and hospitalization data)
- Will help in allocating state resources more effectively

	Prepardness Regions							
Date	PR1	PR2	PR3	PR4	PR5	PR6	PR7	PR8
Jul-20	0.043216	0.287552		0.039519	0.291421	0.058607		
Aug-20	0.083943	0.365514	0.018732	0.104251	0.111955	0.033589		
Sep-20	0.037827	0.425271	0.026686	0.231151	0.377233	0.030864	0.74832	0.286788
Oct-20	0.570891	0.191276	0.300506	0.556949	0.279293	0.369981	0.695218	0.273144
Nov-20	1.257752	1.339831	1.368645	1.952061	1.339907	1.750268	0.969155	1.219505
Dec-20	2.557758	3.729434	1.503731	3.106679	2.280467	2.121647	2.621635	3.573275
Jan-21	3.387304	2.674873	2.08052	2.153733	1.773603	3.658975	1.974418	1.885578
Feb-21	1.173951	2.714585	1.011863	1.283831	1.85568	1.230203	1.135033	1.574897
Mar-21	0.850103	1.006641	0.317984	0.98104	0.820073	0.424564	0.964233	1.595118
Apr-21	1.260708	1.070017	0.303629	0.481643	0.658015	0.301126	0.680736	0.845316
May-21	1.28919	0.147445	0.21914	0.262544	0.571188	0.238763	0.136772	0.174173
Jun-21	0.427957	0.076696	0.060933	0.085263	0.228121	0.163247	0.071689	0.074136
Jul-21	0.306472	0.136311	0.243824	0.203226	0.11787	0.290401	0.50147	0.182568
Aug-21	1.095069	0.149501	0.496198	0.632396	0.443837	0.496582	0.309092	0.321076
Sep-21	0.860178	0.546779	1.047505	0.486416	1.076483	0.830486	0.469958	0.600392
Oct-21	0.900172	0.340904	0.564516	0.430012	0.614339	0.572853	0.284762	0.2075
Nov-21	1.080593	0.773585	1.481117	0.820645	0.983281	0.851582	0.559448	0.451021
Dec-21	1.255118	1.892256	4.202608	1.787222	2.921031	2.298224	0.682347	0.833476

Public Health Response Actions

- State actions taken when increases in viral gene copies are observed:
  - Alerts are emailed to local health districts (LHDs) and utilities.
  - Information is provided on how to interpret the data and link to message toolkit.
  - LHDs are provided case data by sewershed.
  - LHDs are provided with the dashboard containing the raw data and analyses.
- Over **1,500** alerts sent to local health districts and utilities since the network was started to date.

Local Toolkit

Development of toolkit for local health districts and utilities:

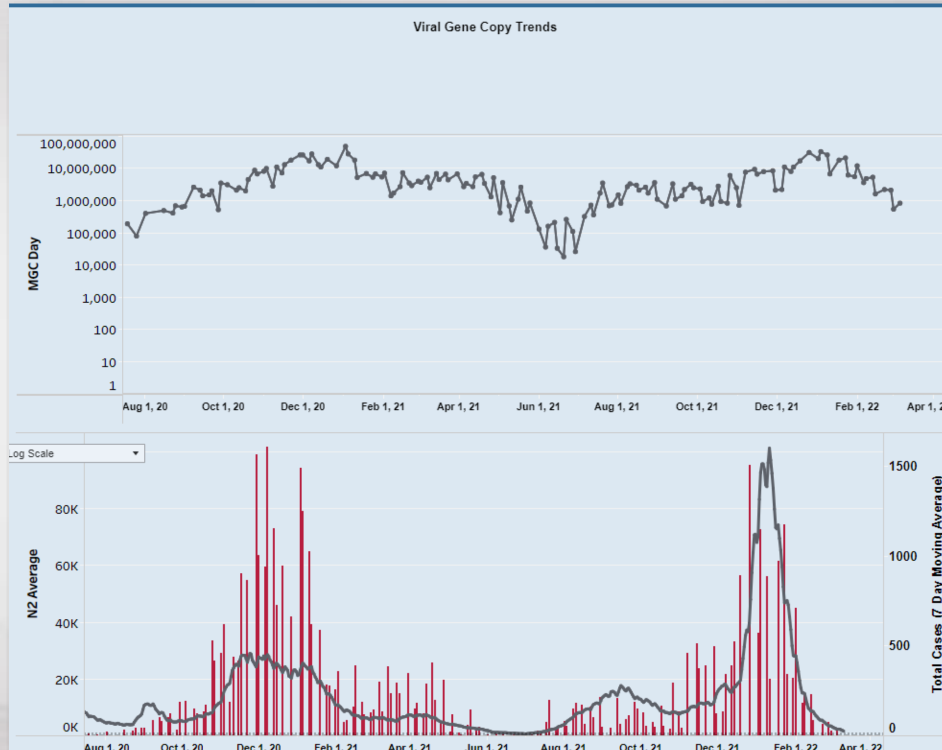
- To serve as an early warning of infection in communities and possibly more targeted areas within communities.
- Additional messaging to public on best practices
- Mobilize additional testing or PPE
- Alert hospitals, physicians, other health care providers
- Closely monitor and evaluate data, (hot spots, contact tracing)
- Provide recommendations to local leaders to take direct actions

Scan the QR code below to see the home page for the ODH Local Toolkit

Ohio Department of Health

Online Dashboard

- Data is presented on the Ohio wastwater monitoring dashboardand the data is uploaded to the Innovate Ohio Platform.
- Nearly 115,000 website hits from August 2020 through July 2021 to the wastewater web page
- CSV download of wastewater and case data by sewershed is available



Download the gene copy data (CSV)   Download the data term definitions (CSV or PDF)   Download information on collection sites (CSV)   Case data by sewershed (CSV)

Participation in NWSS

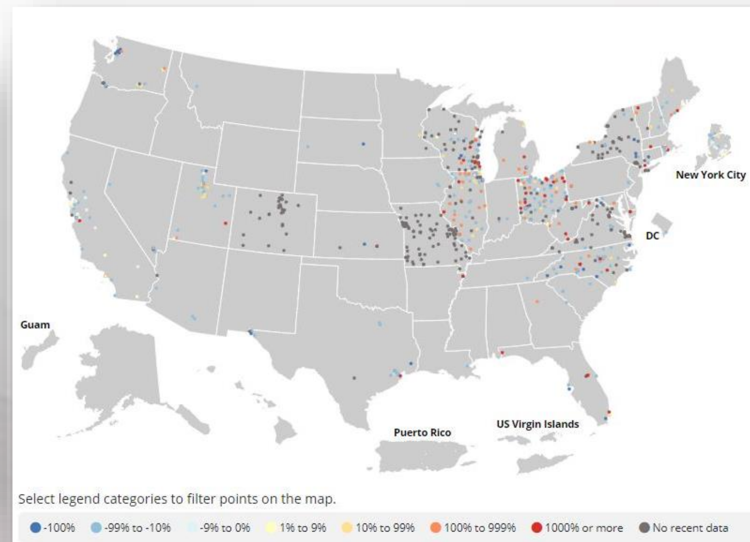
- Ohio was a pilot participant in the CDC National Wastewater Surveillance System (NWSS).
  - CDC has recently released their Covid Data Tracker
- Viral gene copy and case data by sewershed are uploaded daily to NWSS.

SARS-CoV-2 RNA wastewater levels, United States: 15-day percent change by sewershed

15-day % change category	Num. sites	% sites	Category change in last 7 days
-100%	31	7	55%
-99% to -10%	197	47	-33%
-9% to 0%	26	6	8%
1% to 9%	13	3	-7%
10% to 99%	39	9	-37%
100% to 999%	60	14	-6%
1000% or more	53	13	56%

Total sites with current data: 419

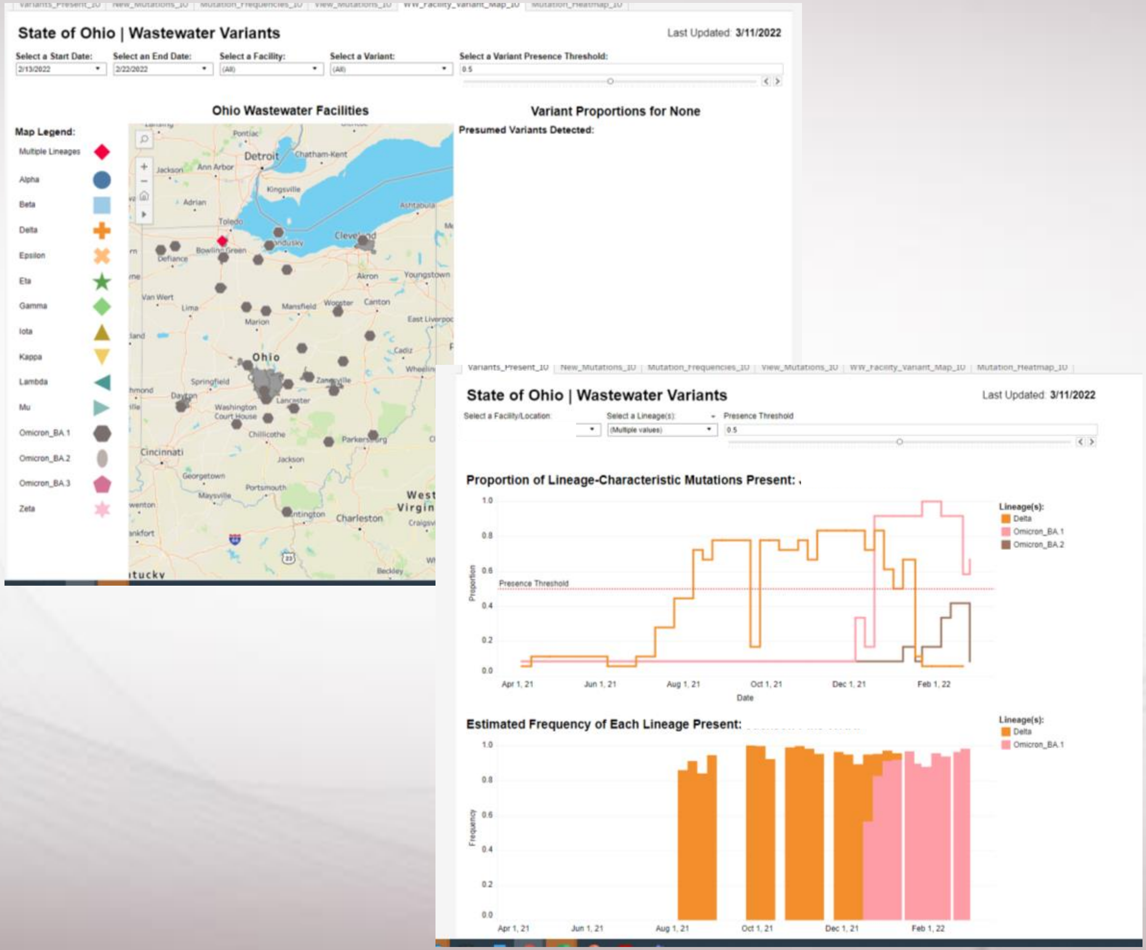
Total number of wastewater sampling sites: 698



https://covid.cdc.gov/covid-data-tracker/#wastewater-surveillance

Wastewater Genomic Sequencing

- Ohio initiated a pilot project with several state universities and US EPA Office of Research and Development in early 2021 to sequence select monitoring sites.
- Weekly sequencing of wastewater samples for all monitoring sites began in April 2021.



Ohio Department of Health

Genomic Sequencing of Wastewater

Goals:

- Use whole genome sequencing data obtained from wastewater to identify the presence of variants and mutations of concern in the absence of, and for comparison to, clinical data.
- Compare wastewater sequencing data to clinical sample data to help inform public health actions.
- Upload and share data to national database.

Challenges:

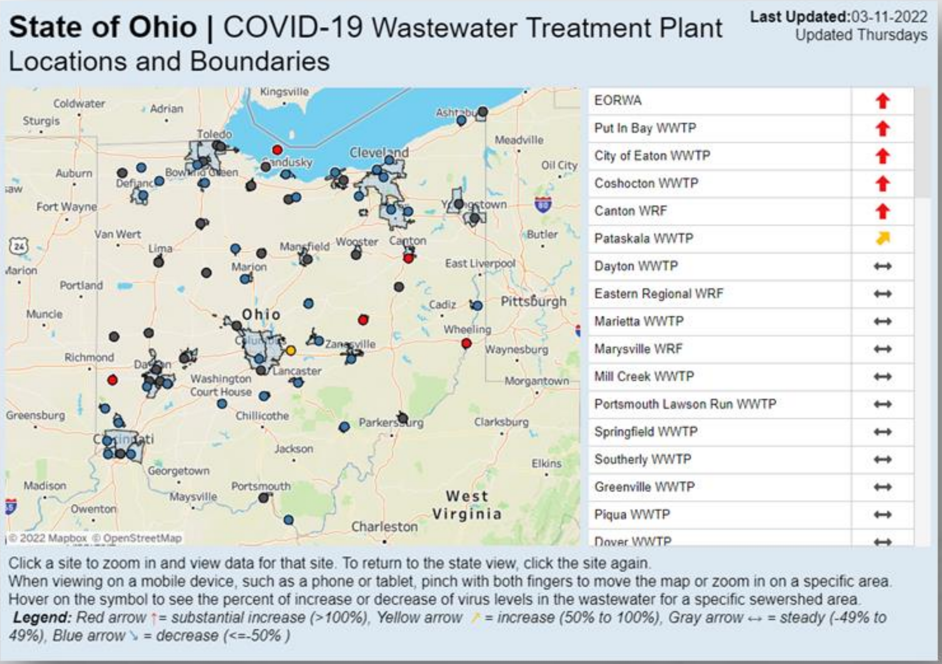
- Wastewater contains fragments of RNA, therefore the consideration of genome coverage, and the occurrence and frequency of mutations associated with specific strains, is important.
- Agreement on mutation list by variant.
- Consistency on how the labs interpret the sequence results.

B.1.1.529.	VOC	E	T9I	Omicron_BA.1
B.1.1.529.	VOC	E	T9I	Omicron_BA.2
B.1.1.529.	VOC	E	T9I	Omicron_BA.3
B.1.1.529.	VOC	M	A63T	Omicron_BA.1
B.1.1.529.	VOC	M	A63T	Omicron_BA.2
B.1.1.529.	VOC	M	A63T	Omicron_BA.3
B.1.1.529.	VOC	M	D3G	Omicron_BA.1
B.1.1.529.	VOC	M	Q19E	Omicron_BA.1
B.1.1.529.	VOC	M	Q19E	Omicron_BA.2
B.1.1.529.	VOC	M	Q19E	Omicron_BA.3
B.1.1.529.	VOC	N	del31/33	Omicron_BA.1
B.1.1.529.	VOC	N	del31/33	Omicron_BA.2
B.1.1.529.	VOC	N	del31/33	Omicron_BA.3
B.1.1.529.	VOC	N	G204R	Omicron_BA.1
B.1.1.529.	VOC	N	G204R	Omicron_BA.2
B.1.1.529.	VOC	N	G204R	Omicron_BA.3
B.1.1.529.	VOC	N	P13L	Omicron_BA.1
B.1.1.529.	VOC	N	P13L	Omicron_BA.2
B.1.1.529.	VOC	N	P13L	Omicron_BA.3
B.1.1.529.	VOC	N	R203K	Omicron_BA.1
B.1.1.529.	VOC	N	R203K	Omicron_BA.2
B.1.1.529.	VOC	N	R203K	Omicron_BA.3
B.1.1.529.	VOC	N	S413R	Omicron_BA.2
B.1.1.529.	VOC	N	S413R	Omicron_BA.3

Future Applications of Wastewater Monitoring

Wastewater monitoring is becoming increasingly vital for community disease surveillance.

- Human testing data for SARS-CoV-2 is decreasing or not reported.
- Less clinical samples of SARS-CoV-2 are available for analysis.
- Ability to monitor other microbiological markers.



Future Applications

- Fall/Winter of 2022
  - Plan to begin monitoring for Seasonal Influenza and Respiratory Syncytial Virus (RSV)

Ohio Wastewater Monitoring Network

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