Wastewater Surveillance of SARS-CoV-2, Influenza, and RSV in Cities, Neighborhoods, and Buildings:

What Works Best and How To Do Better

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COVID-19 trends high and stable in and around Ottawa









Some local vaccine campaigns take steps forward

CBC News · Posted: Oct 12, 2023 12:10 PM EDT | Last Updated: October 12

https://www.cbc.ca/news/canada/ottawa/covid19-ottawa-levels-spread-riskoctober-2023-1.6987460

COVID-19 still present and being accounted for

Belleville, ON, Canada / Quinte News

John Spitters

Oct 12, 2023 | 6:25 AM

https://www.quintenews.com/2023/10/12/covid-19-still-present-and-being-accounted-for/

News > Health

Symptoms of new Covid variants as cases increase in UK

Cases of the Pirola variant in the UK have surpassed 100

https://www.independent.co.uk/news/health/symptoms-of-new-covid-

HEALTH News

A new COVID-19 variant has emerged. Here's what we know about EG.5 so far

https://www.ctvnews.ca/health/a-new-covid-19-variant-has-emerged-here-s-what-we-know-about-eg-5-so-

HPE health unit reports 60 new **COVID-19** cases in region

Published Oct 13, 2023 • 1 minute read

https://www.thewhig.com/news/hpe-health-unitreports-60-new-covid-19-cases-in-region

U.S. to produce more COVID-19 tests for free distribution as cases rise

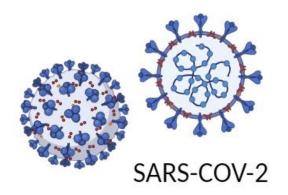


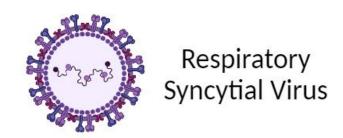
Posted September 21, 2023 8:23 pm · Updated September 21, 2023 8:24 pm

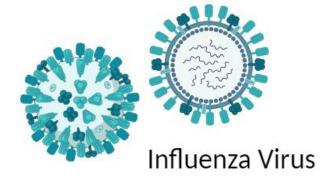
https://globalnews.ca/news/9977423/us-covid-tests-order-canada/



What viruses did we study?







Mpox (Monkeypox) Virus



Wastewater sewer systems are impressive engineering structures!

OTTAWA:

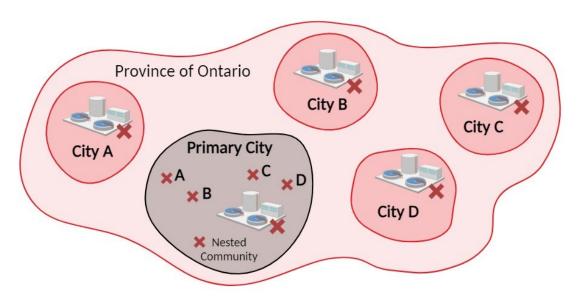
- 2796 km² covered
- 2846 km of sanitary sewers
- 108 km of combined sewers
- 71 wastewater pumping stations
- More than 92,000 manholes
- Approx. 234,000 service connections
- Sewer pipes ranging in size from 20 cm to 3 m in diameter







Where did we sample from?



- → Treatment plant sampling is easy and allows monitoring of larger populations
- → Upper sewershed sampling can provide more specific and actionable information
- Sewer design and hydraulics must be considered in selecting sampling locations
- Going closer to the source reduces residence time in the sewer and viral RNA degradation
- Stormwater, snowmelt, sewer sediment impacts

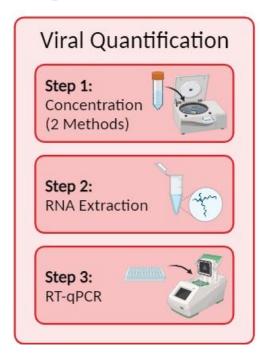


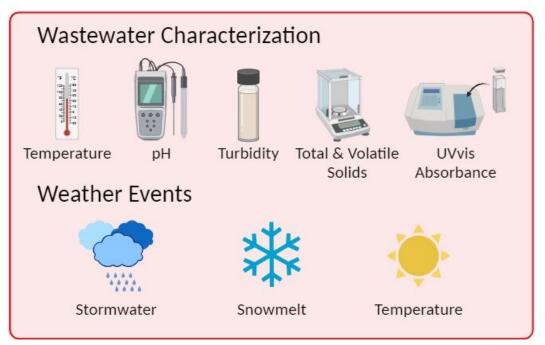
Sewer sampling is not as easy as WWTP sampling!





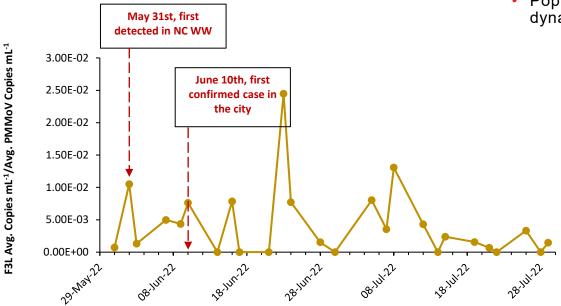
Sample Processing







Approach #1: Nested community compared to City – Monkeypox



Two important criteria in selecting a NC:

- Sewer hydraulics
- Population dynamics/behavior

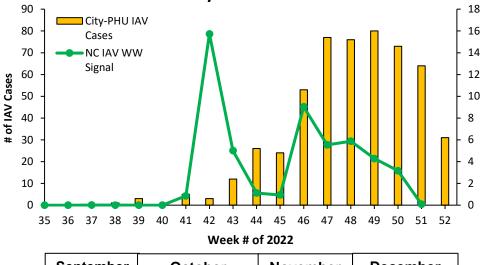


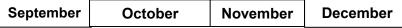
Monkeypox was detected in the nested community wastewater 10 days earlier than the first confirmed case in the City.



Approach #1: Nested community compared to City – Influenza A

Nested-Community IAV WW Levels Compared to the City-PHU IAV Cases





Two important criteria in selecting a NC:

- Sewer hydraulics
- Population dynamics/behavior



Influenza was detected in the nested community wastewater approx. 3 weeks earlier than the rise in the City cases.

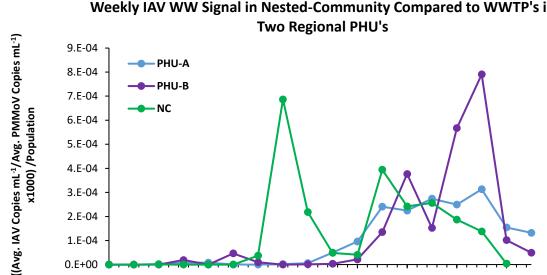
Created in BioRender.com bio



(Avg. IAV Copies mL⁻¹/Avg. PMMoV Copies mL⁻¹)x1000

Approach #2: Nested community compared to the Region - Influenza A

Weekly IAV WW Signal in Nested-Community Compared to WWTP's in



41

0.E+00

35

37

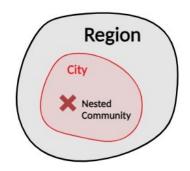
39

Week # of 2022 September October November **December**

43

45

47



Influenza rise was detected in the nested community wastewater approx. 4 weeks earlier than the rise in the PHU-A and PHU-B wastewater and case numbers.

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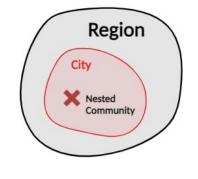


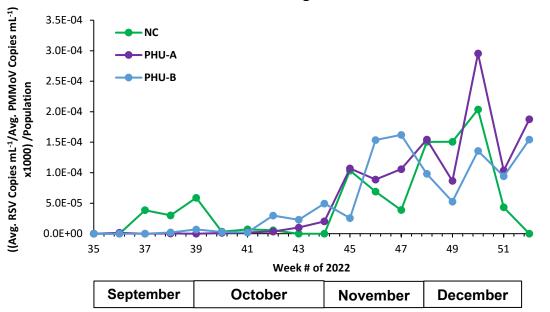
49

51

Approach #2: Nested community compared to the Region – RSV

Weekly RSV WW Signal in Nested-Community Compared to WWTP's in Two Regional PHU's

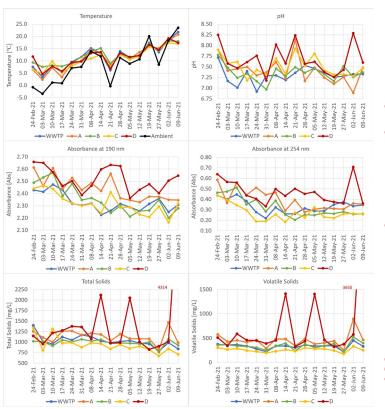


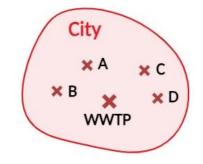


RSV rise was detected in the nested community wastewater approx. 5 weeks earlier than the rise in the PHU-A and PHU-B wastewater.



Approach #3: Sewer Sampling throughout the City

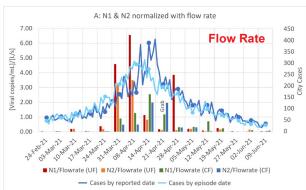


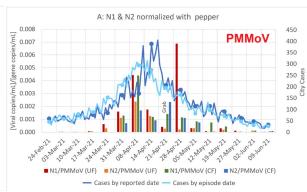


- Strategically selected sampling locations
- Tracked spread of virus and trends in different zones
- Investigated predictive or early indicator ability of sampling locations based on community size, residence times, sewer hydraulics
- Residence times impact wastewater characteristics
- Residence times impact vial RNA signal



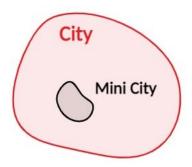
Approach #4: "Mini City" in the City

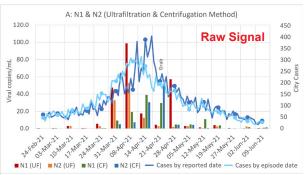














Approach #5: WWTPs in the Region

- 5 cities/towns with varying sewer systems and sizes
- Samples were collected from WWTPs
- SARS-CoV-2 RNA signal in wastewater was correlated to reported and active case numbers
- All sites showed statistical significance (p<0.01 and p<0.05) between viral RNA 3 days prior to case numbers
- Sewer size and combined sewers impacted the significance of correlations
- Normalization with TS, VS, and PMMoV improved correlations
- The best normalization parameter varied from site to site

Created in BioRender.com bic



City D

Province of Ontario

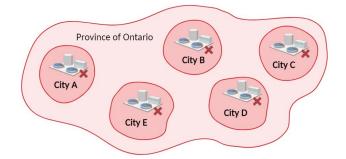
City E

City A

Approach #5: WWTPs in the Region

Impact of External Factors on SARS-CoV-2 Concentrations

Location	TS	VS	Turbidity	Precip.	Snow
Α		**	*	**	*
В		***	**		
С	***	***	***		**
D		***	***	*	*
Е		***	**		
Overall		***	***	**	***



- Statistical analysis to show impacts of external factors
- Viral concentrations impacted by TS, VS, Turbidity, Precipitation and Snow Cover (Dilution effect)

Pearson Correlation Strength: yellow is stronger than green

*p < 0.05, **p < 0.01, ***p < 0.001





Take home messages!

- Upper sewershed and WWTP surveillance are complementary tools in our toolbox
- Sewers are large and very complex structures and impact the viral signal by the time wastewater reaches the treatment plant
- Sewer sampling can provide specific, accurate and actionable information with ample time to prepare and is an early-warning tool
- Sewer hydraulics and design must be considered in selecting sampling locations
- Several approaches can be developed to make the best use of sewershed sampling
- Nested community approach is particularly useful and should be included in wastewater surveillance

Carleton M

Thank You

