

Best Practices for Collection and Storage of Wastewater Samples to Support Wastewater Surveillance of the COVID-19 Signal in Sewersheds

THIS APPENDIX PROVIDES GUIDANCE and consistency in the collection of wastewater samples to support wastewater surveillance of the genetic signal of SARS-CoV-2 in sewersheds. Appropriate sample type, location(s), frequency, duration, etc. will depend on study goals and resources.

Safety

IN ACCORDANCE WITH GUIDANCE from the CDC, standard practices associated with water resource recovery facility operations should be followed, including the PPE normally required when handling untreated wastewater, such as safety gloves and glasses, masks or face shields. Safety recommendations may vary between handling wastewater and processing samples.

Equipment and Preliminary Activities

CLEAN AND PREPARE ALL sample equipment, including composite sampler (if applicable) or sample bucket, 4 x 250 mL polycarbonate leak-proof bottles, cooler with ice, documentation/labels, tubing, and other equipment typically used for measurement of water quality and characteristics.

Sample bottles should be new if possible. The next best alternative is autoclaved bottles, or at minimum, bottles cleaned with bleach and thoroughly rinsed.

Sample Collection

ENSURE SAMPLING DRAWS FROM a well-mixed part of the stream. Sampling near the bottom of a stream may introduce more solids into the sample than what is representative of the entire stream. Collect a sample volume of at least 1 L. Samples may be split in 4 aliquots of 250 mL in order to allow enough sample for analysis, quality control, and archival for future. Do not fill bottles completely, in order to prevent issues upon freezing/storage.

Composite samples are preferred over grab samples, though either is acceptable. Composite samplers need to purge following the collection of each discrete sample to preclude accumulation of solids in sampler tubing. Composite samples should be refrigerated during collection and storage.

If possible, measure additional sample characteristics including air temperature, water temperature, wastewater flow rate, pH, total suspended solids, and chlorine residual.

Documentation

LABEL THE CONTAINERS WITH sample site, date, time, sampler initials, and sample identification number if applicable. Complete the documentation form on the reverse side.

Sample Transport, Storage, and Preservation

SAMPLES SHOULD BE REFRIGERATED during transport, or cooled with ice or cold packs if refrigeration is not available. Once at the laboratory, sample temperature should be recorded, along with other information as documented on the reverse side. Samples should be stored at 4°C and processed as soon as possible (up to a maximum of two weeks). If this is not possible, then samples should immediately be stored at -80°C, or alternatively at -40°C or -20°C (in decreasing order of preference). Avoid freezer defrost cycles.

If possible, it is preferable to store filtered samples rather than raw sewage samples. Methods used for solids removal and concentration should be documented (i.e., filtration, centrifugation, precipitation, skim milk approach). Impacts of pasteurization on the strength of the genetic signal are not known at this time.

Field Sample Collection Form

1. Date field sample collected: _____ Time: _____
2. Type of sample (check one): Grab Composite
If composite, composite type: Flow Time Composite duration: _____
3. Collected by: _____
4. Location (include street, locality, and/or landmarks, as appropriate):

5. Sample ID number/container labeling: _____
6. Collection volume (in mL): _____ Number of aliquots/bottles: _____
7. Wastewater flow rate: _____
8. Did it rain yesterday? Yes No Did it rain today? Yes No
9. Type of sewer system (check one): Separate Combined
10. Air temperature (in degrees Celsius): _____
11. Sample water characteristics: pH: _____ Temperature (in degrees Celsius): _____
Total suspended solids: _____ Chlorine residual: _____
12. Sample transportation/shipping notes:

Other Important Information (If Known)

13. Population served: _____
14. Service area notes: _____
15. Public health data: _____

Storage/Laboratory Information

16. Date sample received: _____ Time: _____
17. Temperature of sample upon receipt (in degrees Celsius): _____
18. Storage temperature (in degrees Celsius): _____
19. Sample processing: _____
20. Preservation agent: _____