

# Standard Operating Procedures for the Collection of Fecal Coliform Bacteria Samples in On-Site Sewage Systems



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## **1. Purpose and Scope**

- 1.1. This document is the Washington State Department of Health (WSDOH) Standard Operating Procedure (SOP) for collecting grab samples directly into sample bottles for the purpose of wastewater sampling and analysis of fecal coliform bacteria. The typical methods for fecal coliform analysis are: Standard Methods (SM) 9222D- a membrane filtration (MF) method and SM9221E the most-probable number (MPN) method.
- 1.2. This SOP includes the procedures for sample collection by hand with an extension sampler pole from a sampling location in an on-site sewage system.

## **2. Applicability**

These procedures are for the proper collection of grab samples for laboratory analysis of fecal coliform. We will collect samples from an on-site sewage system's wastewater stream. This document is for post-UV disinfection unit effluent sampling locations where a free-falling sample can be easily collected by placing a sample bottle under the flow from the end of an open discharge pipe. With a well-mixed wastewater flow stream and with relatively uniform chemistry, these methods described in the SOP are sufficient to represent the effluent of the UV disinfection unit.

## **3. Definitions**

- 3.1. Chain of Custody – An unbroken trail of accountability that assures the physical security of samples, data, and records.
- 3.2. Fecal coliform – A group of bacteria common to the digestive systems of warm-blooded animals that are cultured in standard tests. Counts of these organisms are typically used to indicate potential contamination from sewage or a described level of needed disinfection, generally expressed as colonies per 100 ml.
- 3.3. Field Data Sheet – A weather-resistant standard document containing “Rite in the Rain®” writing paper used to record field activities, sample data, methods and observations for each and all collection sites.
- 3.4. Grab sample – A wastewater quality sample collected during a short time at a single location.
- 3.5. Holding time – The maximum time a sample may be stored prior to its required analysis.
- 3.6. WSDOH - Washington State Department of Health.

## **4. Personnel Qualifications/Responsibilities**

- 4.1. Contracted service providers who are involved in collecting samples must have wastewater sampling experience and/or training to collect representative wastewater samples. Samplers must demonstrate a competency for sample collection using appropriate sampling equipment and techniques. This insures that samplers are equipped with the skill and knowledge to conduct sampling professionally and efficiently.

## **5. Equipment, Reagents, and Supplies**

- 5.1. Clean and sterilized plastic sampling bottles (provided by lab)
- 5.2. Sampling bottle labels and waterproof markers.
- 5.3. Ziploc bags
- 5.4. Extension swing sampler pole
- 5.5. Cooler containing ice (dry ice and freezer packs are not acceptable)
- 5.6. Field data sheet or field data report form (See Appendix B for form.)
- 5.7. Chain of Custody form (See Appendix A for form.)
- 5.8. Site files detailing sampling locations, sample site information, site identification codes
- 5.9. Anti-bacterial hand sanitizer and towels/rags.

- 5.10. Disposable gloves (nitrile or latex)
- 5.11. Face shield or appropriate eye protection.
- 5.12. Tools for removing and securing access lids at sampling locations.

## 6. Summary of Procedures

### 6.1 Direct Post-UV Free-falling Effluent Grab Sampling Procedure

1. Keep the sterile sample bottle unopened until you are ready to collect the sample. Do not rinse the bottle before collection. If the bottle is accidentally contaminated during the sample procedure, replace it with another sterile bottle.
2. Using indelible ink, prepare sample label with the location of sample, sample ID code, sampler's initials, date and time of sample collection.
3. Locate the point downstream from the UV disinfection unit (after UV disinfection of the effluent).
4. Remove the access lid and set aside.
5. Ascertain the ability to collect a representative sample with the extension-sampling pole. Use the following criteria: A) The interior sidewall of the sampling location must provide sufficient working space to allow the open bottle to be withdrawn without contacting the sidewalls and contaminating the sample. B) Proper sampling requires a minimum 4 inches of vertical height between the bottom of the discharge pipe and any structures or liquid level at the sampling location. This provides sufficient room for sample bottle insertion and removal. C) Proceed with sampling after meeting criteria for collection of sample from the free-falling effluent stream at the end of the UV disinfection unit discharge pipe.
6. Secure the sample bottle to the extension-sampling pole clamp. Extend the sampling pole to a length that will allow safe access to the desired sample location.
7. Remove the cap from the sample bottle just before sampling and set it upside down (such as on top of other field equipment). Do not touch or contaminate the underside of the cap, mouth, or the inside of the bottle with your fingers, wind-blown particles, or dripping from your clothes, body, or overhanging structures.
8. Place the opened sample bottle under the free-falling effluent stream. Do not allow the sample bottle mouth to touch the discharge piping or any other source of contamination.
9. When filling the sample bottle, be careful to pull the bottle out of the wastewater stream flow to reach the point at or near the shoulder of the bottle. (The laboratory will provide the sample bottles and instructions of exactly how much to fill them.) If the bottle is filled above this level, then immediately and carefully pour out enough excess sample to ensure that the sample volume is at or near the shoulder (leave at least 1 inch of air space). *Note: This step allows air space for proper mixing at the lab.*
10. Immediately replace the cap securely without touching the mouth of the bottle.
11. Wipe off any dirt, debris, or liquid from the outside of the bottle and attach the completed label.
12. Place the sample bottle in a Ziploc bag to protect the label.
13. Immediately place the sample in a cooler with ice.
14. Place the Chain of Custody (once completed) into a plastic bag and place it inside the cooler.
15. When the sample collection is completed, put the access lid back in place and secure.

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| <p>16. Record the date, time, sample ID code and location with the sampler's name and any other descriptive information pertaining to the sample on the Field Data Sheet. Ensure the bottle label matches the date, time, and sample ID code recorded on the Field Data Sheet.</p> |
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## **7. Storage and Transport**

- 7.1. The sample must be packed in ice and held at 4°C for less than twelve hours (preferably less than six hours) for transport to the testing laboratory. Re-freezable ice packs are not sufficient to ensure compliance with the required holding temperature and are prohibited. Use insulated containers to assure proper maintenance of storage temperature. Ensure that sample bottles are not totally immersed in water during transit

## **8. Records Management**

- 8.1. The service provider lead will keep the completed Field Data Report Form and any other hard-copy forms. Organize these documents in binders or expanding files.

## **9. Field Data Sheets**

- 9.1. Complete a field data sheet (FDS) for each sample collected. The FDS details specific observations pertaining to each sample. Required information recorded on the FDS includes.
- Date, arrival time, departure time.
  - Name of person and company collecting the sample.
  - Sample site address and sample ID code.
  - Specific sampling location within treatment train.
  - Sampling time and date.
  - Environmental conditions at each sampling location.
  - Pertinent observations and/or any problems with sampling, including deviations from sampling procedures.
  - Unusual circumstances that might affect interpretation of results.

## **10. Chain of Custody**

- 10.1. Written records must accurately trace the custody of each sample collected through all phases of the UV disinfection-unit field study. The primary objective of a chain of custody is to create an accurate written record to trace the possession and handling of the sample from the moment of its collection to final analysis. Use the chain of custody form provided in Appendix A. Turn in a chain of custody form to the laboratory with every sample delivery.

## **11. Quality Control and Quality Assurance Section**

- 11.1. The data QA program for field sampling consists of two parts: 1) adherence to the SOP procedures for sample/data collection and periodic evaluation of sampling personnel and 2) the collection of a field quality control (QC) sample for 10% of regular samples collected. The field QC sample is collected as a replicate sample immediately after the regular sample at the same location. Precision or variability for the field sampling is assessed by collecting one replicate sample at the rate of 10% of regular samples collected.
- 11.2. Submit the replicate samples to the laboratory labeled as a QC sample. The lab should treat the sample in the same way as a regular field sample.

## 12. Safety

- 12.1. Always wear appropriate eye protection, waterproof disposable gloves and protective clothing. Must use new, clean disposable gloves at each sampling location.
- 12.2. Take care to disinfect equipment, dispose of gloves properly, and wash hands thoroughly after collection of sample.
- 12.3. Avoid touching face, mouth, eyes, or nose before washing hands.
- 12.4. Always avoid eating, drinking, or smoking near samples, and at the sampling location.
- 12.5. Existing scrapes, cuts, and burns must be covered whenever there is potential contact with wastewater. Promptly treat new cuts and abrasions, including those that are minor, using appropriate first aid measures.
- 12.6. When using equipment in the field be aware of your surroundings. Select an area in which you feel safe and secure from hazards.

## 13. References

American Public Health Association (APHA) (2012). *Standard Methods for the Examination of Water and Wastewater*. 22<sup>st</sup> ed. E.W. Rice, R.B. Baird, A.D. Eaton, and L.S. Clesceri, eds. APHA-AWWA-WEF, Washington D.C.

Eliasson, John M. 1997. Wastewater Sampling Protocols for On-Site Sewage Systems. 9<sup>th</sup> Northwest On-Site Wastewater Treatment Short Course and Equipment Exhibition.

Norweco, Inc. Effluent Sampling For Residential Treatment Systems.

Washington State Department of Ecology. 2011. Standard Operating Procedures for the Collection of Fecal Coliform Bacteria Samples in Surface Water: Version 2.1 Environmental Assessment Program.

Washington State Department of Ecology. 2009. Standard Operating Procedures for Collecting Grab Samples from Stormwater Discharges: Version 1.0 Environmental Assessment Program.

**Appendix A – Chain of Custody Form (sample)**

**Project Name**  
UV Disinfection Study

**Sampler**  
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**Company Name**  
Wastewater Management  
Office of Environmental Health and Safety  
Washington State Department of Health

**Company Address and Phone**  
243 Israel Road SE  
Tumwater, WA 98501  
(360)236-3379

Sample ID	Date	Time	Location	Type (Normal/ Quality Control)	Preservative Method	Temp at Delivery (°C)
					ice	
					ice	
					ice	
					ice	
					ice	
					ice	
					ice	
					ice	
					ice	
					ice	
					ice	
					ice	
					ice	
					ice	
					ice	
					ice	

All samples are grab samples. All samples delivered by hand to lab.

Relinquished by: <i>(Printed name and signature)</i>			Received by: <i>(Printed name and signature)</i>		Date	Time
Relinquished by: <i>(Printed name and signature)</i>			Received by: <i>(Printed name and signature)</i>		Date	Time
Relinquished to lab by: <i>(Printed name and signature)</i>	Date	Time	Received for lab by: <i>(Printed name and signature)</i>		Date	Time

## Appendix B - Field Data Report Form



### FIELD DATA LOG UV Disinfection Unit

Site address: \_\_\_\_\_

Evaluation completed by: \_\_\_\_\_

Evaluation completed on: Date: \_\_\_\_ / \_\_\_\_ / \_\_\_\_

Arrival Time: \_\_\_\_\_

Departure Time: \_\_\_\_\_

Sampling Time: \_\_\_\_\_

Sampling Location:

Ambient Temperature: \_\_\_\_\_ °C

Barometric Pressure: \_\_\_\_\_ in Hg

Parameter	Value	Unit	Comments
UV Transmittance		%	
Turbidity		NTU	
Dissolved Oxygen		mg/L	
pH		units	
Conductivity		μS/cm	
Temperature		°C	

Other notes (weather conditions, wastewater characteristics that may compromise treatment):