Indicator Bacteria in Fresh Water

The following tables are not applicable to marine water samples.

A list of species included in this category may be found in the associated QAPrPTableReference.

Terms appearing in the tables are defined in the *Surface Water Ambient Monitoring Program Quality Assurance Program Plan*, which contains a glossary (Appendix E), as well as a list of abbreviations and acronyms (Appendix F).

Table 1: Quality Control[1]: Indicator Bacteria in Fresh Water

Laboratory Quality Control	Frequency of Analysis	Measurement Quality Objective
Sterility Checks[3]	Per new lot of dehydrated culture media as instructed in SM 9020B.4.i.5[2] and SM 9222D.1.a	No growth
	For non-sterile filters and pads per lot as instructed in SM 9020B.4.h.1.1	No growth
	Membrane Filter Media, filters, buffered dilution water, rinse water, and all equipment per series of samples as instructed in SM 9020B.8.a.5[2]	No growth
	Multiple Tube Media, dilution water, and glassware as instructed in SM 9020B.8.a.5[2]	No growth
Laboratory Positive Control	Per new lot of dehydrated culture media for the following methods: Colilert, Colilert -18, Colisure, Enterolert, or other chromogenic/fluorogenic methods. Per new lot of commercially-prepared culture media ampules for USEPA-approved fecal coliform and E. coli membrane filter methods (e.g. SM 9222, m-ColiBlue24, EPA 1603) Per batch for laboratory-prepared culture media for USEPA-approved fecal coliform and E. coli membrane filter methods (e.g., SM 9222)	Positive response
Laboratory Negative Control	Per new lot of dehydrated culture media for the following methods: Colilert, Colilert -18, Colisure, Enterolert, or other chromogenic/fluorogenic methods. Per new lot of commercially-prepared culture media ampules for USEPA-approved fecal coliform and E. coli membrane filter methods (e.g. SM 9222, m-ColiBlue24, EPA 1603) Per batch for laboratory-prepared culture media for USEPA-approved fecal coliform and E. coli membrane filter methods (e.g., SM 9222)	Negative response
Laboratory Duplicate	Per 10 samples or per analytical batch, whichever is more frequent	$R_{log} \leq 3.27 \times \overline{R}^{4}$ Computation of R from duplicate laboratory sample analyses
Laboratory Blank[5]	Per 10 samples or per analytical batch, whichever is more frequent	No growth

Field Quality Control[6]	Frequency of Analysis	Measurement Quality Objective
Field Blank, Equipment Blank	Per method or SOP	Negative response

[1] Unless method specifies more stringent requirements

[2] Citations from *Standard Methods for the Examination of Water and Wastewater*, 20th edition(4)

[3] Sterility Checks

The specific type and number of sterility checks are method-dependent. For example, membrane filter tests require the testing of filters for sterility, while multiple-tube or pour plate procedures do not.

[4] Method for Determining Precision

In order to determine precision for bacterial analysis, the following procedure (adapted from Standard Methods 9020 Section 8.b) will be used. Note: When determining the precision of bacterial analyses, it is important to distinguish between different matrices (drinking water, wastewater, ambient water). Duplicate results from different matrices must be kept separate when calculating precision.

In order to calculate the laboratory precision for bacterial analyses, the results from the preceding 15 positive samples of a specific type (matrix) are used to calculate a running mean. The results used to calculate the running mean must all correspond to the same quality control parameter, in this instance laboratory duplicates (as opposed to field duplicates). The results of different quality control parameters such as laboratory and field duplicates must not both be used to calculate a single running mean. Note: Field duplicates are not a current SWAMP requirement (see footnote 6).

Step 1: Record the results from duplicate analyses (these results are here designated as D_1 and D_2).

Step 2: Calculate the logarithm (here designated as L_1 and L_2) of each duplicate result. Note: If either of the values D_1 or D_2 are less than 1, add 1 to both values before calculating the logarithms.

$$L_1 = \log D_1$$
$$L_2 = \log D_2$$

Step 3: Calculate the range of logarithms (R_{log}) for each pair of duplicates. R_{log} is equal to the absolute value of the difference between the two numbers.

$$R_{log} = |L_1 - L_2|$$

Step 4: Calculate the mean of R_{log} ($\overline{R}$) for the duplicates analyzed

$$\overline{R} = \sum \frac{R_{log}}{n}$$

Where

$\sum R_{log}$ = the sum of the ranges of logarithms calculated for each pair of duplicates
n = the number of pairs of duplicates (in this case, n = 15)

Step 5: Assess the precision of the duplicate analyses. In order for the laboratory to demonstrate an acceptable level of precision, the range of logarithms for a particular duplicate must be less than the mean of the range of logarithms multiplied by 3.27.

$$R_{log} \leq 3.27 \times \overline{R}$$

[5] Laboratory Blanks

Analysis and reporting of laboratory blanks is required only when samples are diluted prior to analysis. If samples are not diluted in the sample batch, no laboratory blanks are required for that specific sample batch.

[6] Field Duplicates

While SWAMP recommends that field duplicates be collected and analyzed, they are not a current SWAMP requirement. Projects are encouraged to require field duplicates in their QA project plan (QAPP) if it supports their specific quality objectives.

Table 2: Sample Handling: Indicator Bacteria in Fresh Water

Recommended Container	Recommended Preservation	Required Holding Time[1,2]
Factory-sealed, pre-sterilized, disposable whirlpak bags or 125-mL sterile plastic (high density polyethylene, polystyrene, or polypropylene) or glass container	Cool to ≤10 °C; for samples containing chlorine, sodium thiosulfate is pre-added to the containers in the laboratory	8 hours *for compliance monitoring*
		24 hours *for routine ambient monitoring*

[1] Each "Required Holding Time" is based on the assumption that the "Recommended Preservation" (or a method-mandated alternative) has been employed. All samples analyzed past the 8 hour compliance holding time will be flagged for user notification, however, will still be considered SWAMP compliant for routine ambient use. If the 24 hour holding time for analysis is not met, the project manager and SWAMP Quality Assurance Officer must be notified and the data must be flagged accordingly.

[2] Sample analysis should begin as soon as possible after receipt, a holding time of no more than 8 hours is highly recommended. For purposes of compliance monitoring, sample incubation must be started no later than 8 hours from time of collection and no later than 24 hours for routine ambient monitoring. (1, 2, 3, 4)

Table 3: Corrective Action: Indicator Bacteria in Fresh Water

Laboratory Quality Control	Corrective Action
Sterility Checks	Identify contamination source and take appropriate action; discard membrane filter/pad or prepared media lot; discard sample results if checks made during analysis
Laboratory Positive Control	Identify cause and take appropriate action; discard prepared media and remake from start or purchase new lot
Laboratory Negative Control	Identify cause and take appropriate action; discard prepared media and remake from start or purchase new lot
Diluent Control	Identify contamination source and take appropriate action; qualify data as needed
Laboratory Duplicate	Verify results; qualify data as appropriate
Laboratory Blank	Identify contamination source and take appropriate action; qualify data as needed
Field Quality Control	**Corrective Action**
Field Blank, Equipment Blank	Examine field log; identify potential contamination source; qualify data as needed

References:

(1) Meyers, D.N., et. al. 2014. U.S. Geological Survey TWRI Book 9. Fecal Indicator Bacteria. Ch. 7, V. 2.

(2) Pope, M.L., et. al. 2003. Assessment of the Effects of Holding Time and Temperature on *Escherichia coli* Densities in Surface Water Samples. Applied and Environmental Microbiology, Vol. 69, No. 10, p. 6201-6207.

(3) Standard Methods Committee. SM Section 9060. Standard Methods for the Examination of Water and Wastewater. Version 2006.

(4) Standard Methods Committee. Standard Methods for the Examination of Water and Wastewater, 20th Edition.