Disclaimer: The information in this guidance document does not supplant to provisions of the Environmental Laboratory Accreditation Regulations, 25 Pa Code, Chapter 252 or the TNI Standard. This document is a tool to help laboratories comply with Chapter 252 and the TNI Standard. If there is any disagreement between the contents of this document and Chapter 252 and the TNI Standard, the regulations or standard shall prevail. This document outlines the Department of Environmental Protection's ("Department") Laboratory Accreditation Program ("LAP") of the Department's expectations regarding the reporting and notification requirements for microbiological testing of SDWA compliance samples. Specifically, the analysis dates and times, validation and invalidation of results and public notification.

Analysis Dates and Times:

The "analysis date and time" for the purposes of compliance with the Environmental Laboratory Accreditation Regulations (25 Pa. Code Chapter 252) is the date and time when the sample is placed into the incubator after filtration or preparation. For the purposes of this document please refer to this as the "accreditation analysis date and time." This means that for a sample to be analyzed within the holding time the sample must be placed into the incubator before the expiration of the holding time. The accreditation analysis date and time would be the date and time that must be reported on a final analytical test report to a client.

The "analysis date and time" for the purposes of compliance with reporting results to the Department's "Drinking Water Electronic Laboratory Reporting" ("DWELR") system and the PA Safe Drinking Water Regulations (25 Pa. Code Chapter 109) is the date (and time for public notification purposes) when the final analytical result is obtained. In the case of microbiology results, this date (and time) would be when the samples are taken out of the incubator and the samples are "read" by the analyst to determine presence, absence, or counts. For presence/absence samples performed by methods with presumptive and confirmatory steps (such as SM 9222B + 9222G and SM 9221B + 9221F) , this date (and time) is the date (and time) the confirmation samples are taken out of the incubator and the samples are read by the analyst. Presumptive positive results should not be reported to DWELR before the confirmation steps are completed. For the purposes of this document please refer to this as the "DWELR analysis date and time."

Validation and Invalidation of Results:

The laboratory is responsible for verifying sample acceptance before sample analysis begins. The validation of sample acceptance includes ensuring that samples are:

- collected in the proper container with the correct volume,
- properly preserved (which includes removal of residual chlorine),
- properly stored, and
- analyzed within the holding time.

The laboratory should reject any sample that does not meet the sample acceptance criterion. If the laboratory chooses to accept samples that do not meet the sample acceptance criterion and the analysis of samples occurs when any of the above criteria are not met, the laboratory cannot invalidate a positive sample result without approval from the Department's LAP.

The validation of positive microbiological results is very simple. Only the Department may invalidate positive microbiological test results.

- If a laboratory accepts samples and begins the analysis by placing the samples in the incubator, if a positive result is observed, the positive sample result is considered valid.
- Even if the samples were not incubated at the correct temperature, for the correct amount of time, a blank is positive, the samples were analyzed out of hold, etc. the laboratory must consider the positive coliform result as "valid." These results trigger the public notification requirements of §§ 109.810(b)(1) and (2).

The validation of negative microbiological results is more complicated.

- In the case of incubation time, samples that are not incubated long enough or for some methods are incubated too long cannot be deemed "negative" and are therefore not valid for compliance purposes. For samples that are incubated too long, the laboratory should consult with the LAP to determine validity of the negative results.
- In the case of incubation temperature, samples that are incubated at temperatures outside the method specified temperature range cannot be deemed "negative" and are therefore not valid for compliance purposes.
- In the case of potential contamination issues (such as media and supply sterility checks, cross-contamination during membrane filtration, etc.), the laboratory must coordinate with the LAP to determine if the "negative" sample results can be considered valid for compliance purposes.
- In the following cases, as described in § 109.301(3)(iii)(B), the laboratory shall invalidate a "negative" coliform result for the following cases:
    - The sample produces a turbid culture in the absence of gas production using an analytical method where gas formation is examined.
    - The sample exhibits confluent growth or produces colonies too numerous to count ("TNTC") with an analytical method using membrane filtration.

25 Pa. Code Chapter 252, § 252.708(a)(2) states that microbiological samples results must be reviewed within 24 hours of acquisition of the initial sample results and the 24 hour deadline may be extended to a maximum of 72 hours to accommodate a holiday or weekend when the laboratory is closed for business. As described in the "Validation and Invalidation of Results" section above, all positive microbiological sample results are automatically considered valid upon being "read" by the analyst, also known as the DWELR analysis date and time. Therefore, it is not appropriate to wait 24 or 27 hours from the initial sample reading to begin the public notification requirements of §§ 109.810(b)(1) and (2).

Public Notification:

For microbiological testing, as soon as samples are placed in the incubator, water bath, or upon the start of the testing procedure for tests that do not require incubation, the laboratory assumes the responsibility for reporting any positive coliform results in accordance with the public notification requirements of 25 Pa. Code Chapter 109, §§ 109.810(b)(1) and (2). This means that regardless of the sample acceptance criteria, performance of the associated QC (e.g.: sterility blanks), instrument performance (e.g.: incubator temperatures), time of incubation, etc., the laboratory MUST report any and all positive coliform results in accordance with §§ 109.810(b)(1) and (2).