

**LABORATORY QUALITY ASSURANCE EVALUATION PROGRAM FOR
ANALYSIS OF *CRYPTOSPORIDIUM* UNDER THE SAFE DRINKING
WATER ACT**

Information Collection Request:

Supporting Statement

**U.S. ENVIRONMENTAL PROTECTION AGENCY
Office of Ground Water and Drinking Water**

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Information Collection Request Section 1: Part A of the Supporting Statement

1. Identification of the Information Collection

1(a) Title of the Information Collection

Laboratory Quality Assurance Evaluation Program for Analysis of *Cryptosporidium* under the Safe Drinking Water Act (Renewal)

OMB Number: 2040 - 0246
U.S. EPA Tracking Number: 2067.04

1(b) Short Characterization

The U.S. Environmental Protection Agency (EPA) is requesting a renewal of the information collection request (ICR) for the Laboratory Quality Assurance Evaluation Program for Analysis of *Cryptosporidium* under the Safe Drinking Water Act (Lab QA Program). This voluntary program applies to public and private laboratories that analyze water samples for *Cryptosporidium*. The program will help ensure that laboratories meet the quality assurance and the updated quality control criteria in Appendix J when using EPA Methods 1622 (EPA-815-R-05-001, <http://www.epa.gov/nerlcwww/documents/1622de05.pdf>) or 1623 (EPA-815-R-05-002, <http://www.epa.gov/nerlcwww/documents/1623de05.pdf>) for determining the identity and concentration of *Cryptosporidium* in source water by filtration, immunomagnetic separation (IMS), and immunofluorescence assay (IFA) microscopy. EPA Methods 1622 and 1623 were designated for *Cryptosporidium* analyses in the LT2ESWTR (40 CFR 141.704); EPA Method 1623.1 (EPA 816-R-12-001, <http://water.epa.gov/scitech/drinkingwater/labcert/upload/epa816r12001.pdf>) was approved as another option for LT2ESWTR analyses per the June 28, 2012 Federal Register Notice, "Expedited Approval of Alternative Test Procedures for the Analysis of Contaminants Under the Safe Drinking Water Act" (<http://water.epa.gov/drink/resources/new.cfm>).

Information collection activities required for laboratories seeking State Certification include: laboratory documents with resumes for each staff member; standard operating procedures (SOPs); Ongoing Precision and Recovery (OPR) control charts; Matrix Spike control charts; training records for all analysts/technicians; proficiency testing (PT) results; and a copy of any certificates as documentation of equipment maintenance; participating in off-site evaluation activities, including submitting positive staining control and OPR slides and performing online analyst evaluation; and participating in a 1 to 2 day on-site evaluation of laboratory performance and data quality, including performing an independent count of a blind slide. All materials are being collected by participating State certification programs. This information collection will provide States with data to verify that the laboratories are capable of producing reliable data from the analysis of *Cryptosporidium* using EPA Method 1622, EPA Method 1623 or EPA Method 1623.1.

The information collection will involve approximately 45 laboratories at a total cost of approximately \$590,858.79 or 3504 labor hours annually. The total number of laboratories includes 45 laboratories that participate in the proficiency testing program. The estimated total State burden, including contractual costs, is estimated at \$212,916.00 or 1968 labor hours annually, based on an estimate of 20 participating States (Appendix K). The estimated total Agency burden, including contractual costs, is estimated at \$125,170.15 or 1953 labor hours annually (Appendix K).

2. Need For and Use of the Collection

2(a) Need/Authority for the Collection

The information collection is needed by EPA to support the *Cryptosporidium* data gathering activities that are required under the Long Term 2 Enhanced Surface Water Treatment Rule (LT2ESWTR). The Lab QA Program is being renewed because the *Cryptosporidium* laboratory evaluation program must continue for the duration of the LT2ESWTR. Renewing the Lab QA Program will help ensure that qualified laboratories are available to public water systems.

2(b) Practical Utility/Users of the Data

Information collected under the Lab QA Program will be used by States and EPA to verify that *Cryptosporidium* occurrence data are generated by qualified laboratories that can perform the analyses acceptably. Use of qualified laboratories for source water monitoring by drinking water utilities will help ensure that the data collected are of known and reliable quality. Data quality could potentially be compromised in the absence of Laboratory Certification Programs.

A list of laboratories that meet the evaluation program criteria will be made available to the public by States and/or EPA at http://water.epa.gov/lawsregs/rulesregs/sdwa/lt2/lab_home.cfm and will provide a resource to aid drinking water utilities (and others interested in monitoring water for *Cryptosporidium* occurrence for the protection of public health) in selecting a qualified analytical laboratory.

3. Non-duplication, Consultations, and Other Collection Criteria

3(a) Non-duplication

The information requested from the respondents under this ICR is not available from other sources. The information requested will be used to assess the current ability of a laboratory to reliably analyze *Cryptosporidium* in water using EPA Method 1622, EPA Method 1623 or EPA Method 1623.1. Information submitted for previous programs, such as the Information Collection Rule, is not applicable because older analytical methods were used and quality control requirements were different. The determination that this information is not available from other sources was made by the Office of Ground Water and Drinking Water Technical Support Center

(TSC), which will be administering the Lab QA Program, and which has worked closely since 1996 with the capable laboratories that will be affected by this information collection.

3(b) Public Notice Required Prior to ICR Submission to Office of Management and Budget

On September 5, 2013 (78 FR 54643), EPA sought comments on this ICR pursuant to 5 CFR 1320.8(d). EPA received two comments during the comment period, which are addressed in the ICR. Copies of the previous four Federal Register notices are attached in Appendices A, B, C, and D. The ICR was last renewed in 2009 (See Appendix D). EPA has developed a webpage to provide further information on the program. The website can be accessed at http://water.epa.gov/lawsregs/rulesregs/sdwa/lt2/lab_home.cfm.

3(c) Consultations

EPA conducted meetings with representatives of the drinking water treatment industry and the community of laboratories expected to seek EPA recognition under the Laboratory Quality Assurance Evaluation Program in Cincinnati, OH, on January 23 and March 12-13, 2001, and in Washington, DC, on February 13-14, 2001. EPA presented and discussed draft plans for the laboratory evaluation program at these meetings and sought input from the drinking water utility and laboratory representatives that attended these meetings. In addition, EPA has been working with States and EPA Regions to integrate *Cryptosporidium* laboratory approval/oversight into existing State certification/accreditation programs. Participating States will approve and oversee laboratories that support the second round of LT2ESWTR monitoring.

Laboratories were asked to estimate the loaded cost of legal, clerical, technical and management personnel per hour, and average cost for analyses of a sample. In addition, laboratories were asked to estimate the hours that each category of personnel would spend to complete the activities associated with submitting a laboratory evaluation application package, performing off-site evaluation activities, preparing and hosting on-site evaluation, and performing and reporting two sets of PT samples per year. Laboratories were also asked to estimate the operation and maintenance costs associated with PT samples and audits. Six laboratories have supplied burden estimate information for participating in a state laboratory certification program:

- Charlotte-Mecklenberg Utilities – Environmental Laboratory Services
- Pennsylvania Department of Environmental Protection, Bureau of Laboratories
- Environmental Science Corporation
- BioVir Laboratories, Inc.
- CH Diagnostic & Consulting Service, Inc.
- Underwriters Laboratories, Inc.

States were asked to estimate the loaded cost of legal, clerical, expert, technical or certification officer, and management personnel per hour. In addition, states were asked to estimate the hours that each category of personnel would need to complete the activities associated with reviewing a laboratory evaluation application package, conducting off-site evaluation activities, conducting on-site evaluations, notifying laboratories of certification status,

reviewing and tracking PT samples, coordinating follow-up activities for failed PT results and maintaining a list of certified laboratories on the State website. Two states have supplied burden estimates for providing laboratory certification:

- New Jersey Department of Environmental Protection, Office of Quality Assurance
- Kentucky Department of Environmental Protection, Division of Water

During the public comment period announced in the September 5, 2013 Federal Register Notice, EPA received two sets of comments – one from a commercial laboratory and another from the American Water Works Association (AWWA). A number of the comments address matters beyond the scope of this ICR and will be addressed separately. Several of the comments pertain to the scope of the ICR and were addressed as follows.

AWWA asked that EPA “Advise [PWSs] on the status and timeline for the current lab approval process... by 12/31/13 and provide a regularly updated, centralized list of all approved labs and the states that approved each.” Laboratories are being approved to support Round 2 monitoring on an ongoing basis. The latest information regarding laboratory approval status is posted on the EPA Website http://water.epa.gov/lawsregs/rulesregs/sdwa/lt2/lab_home.cfm. EPA’s Technical Support Center will maintain a list of links to State Web sites naming certified laboratories as well as the list of certified laboratories on EPA’s website.

AWWA commented that there was a “Lack of information upon which water systems can organize and budget procurement of (qualified lab) services.” A complete section on “Contracting for Laboratory Services” is included in the Public Water System Guidance Manual for the Long Term 2 Enhanced Surface Water Treatment Rule (LT2 Rule). This guidance manual for PWSs affected by the rule provides information on laboratory contracting, sample collection procedures, and data evaluation and interpretation advice available at <http://www.epa.gov/safewater/lt2/index.html>. Responses to frequently asked questions (FAQs) on sampling, analysis, and data reporting questions for the LT2 rule also are available on this website.

AWWA also suggested that “EPA cannot require use of EPA Method 1623.1 or change the ongoing precision and recovery (OPR) requirements for EPA Methods 1622 or 1623 without rule making...” The Agency approved Method 1623.1 as another option for LT2ESWTR analyses pursuant to the June 28, 2012 Federal Register Notice, “Expedited Approval of Alternative Test Procedures for the Analysis of Contaminants under the Safe Drinking Water Act.” EPA has also used this ICR action to provide notice of an update of the 2009 laboratory quality assurance procedures for Methods 1622 and 1623 to reflect that improved recovery for *Cryptosporidium* in ongoing precision and recovery (OPR) samples is now routinely achievable as labs have gained experience with *Cryptosporidium* measurement. Minimum recovery is based on an updated and expanded data set from laboratories using Method 1623 and should provide a better assessment of laboratory performance than the previous value for the following reasons: 1) the data set is more recent; and 2) the sample size is more than twice as large as the 2009 sample size.

Lastly, both the commercial laboratory and AWWA voiced concern regarding the

increased costs for lab approval. EPA notes that the approach for Round 2 creates consistency with the approach for approval/oversight of laboratories for all other regulated analytes and represents an opportunity for greater efficiencies as the oversight for *Cryptosporidium* is integrated into existing programs. EPA believes that the net impact of this approach on analytical costs will be marginal as any fees imposed by certification bodies are distributed across the laboratory analyses. For specific burden breakdowns, refer to the Participating Laboratories Seeking Certification and State Annual Burden tables in Appendix K of this Supporting Statement.

3(d) Effects of Less Frequent Collections

Under the Lab QA Program, laboratories analyze single-blind proficiency test samples 2 times per year. This frequency enables States to independently verify that laboratories perform in an acceptable manner. Less frequent proficiency test samples may not sufficiently capture a laboratory's performance over time. Laboratories report proficiency test results to the PT provider within 14 days of sample receipt. Reporting proficiency test sample results at this frequency allows States to respond in a timely manner to any problems the laboratory may be having with analysis of *Cryptosporidium* in water.

3(e) General Guidelines

The Lab QA Program adheres to all of OMB's general guidelines for information collection.

3(f/g) Confidentiality/Sensitive Questions

The Lab QA Program does not ask any confidential or sensitive questions.

4. The Respondents and the Information Requested

4(a) Respondents/NAICS Codes

The following is a list of NAICS codes associated with laboratories affected by the requirements of this ICR:

541380 – Professional, Scientific, and Technical Services: Testing Laboratories

924110 – Public Administration: Administration of Air and Water Resources and Solid Waste Management Programs

4(b) Information Requested

(i) Data Items

Data items requested from laboratories:

- Laboratory evaluation application information
- Resumes for each staff member seeking EPA recognition under the program

- Standard operating procedures (SOPs)
- Ongoing precision and recovery (OPR) and matrix spike (MS) control charts
- Training records for all analysts/technicians added since the last audit
- Data packages
- Documentation of off-site evaluation activities, including submitting positive control and OPR slides and performing internet analyst evaluation
- Proficiency testing (PT) data
- Documentation of corrective actions taken in response to any deficiencies noted during the on-site evaluation
- Documentation of corrective actions taken in response to poor PT results

Laboratories and States maintain:

- Proficiency Test data

(ii) Respondent Activities

Activities for laboratories seeking state certification:

- Completing laboratory evaluation application package (no more than 1 time per 2 year period) (See Appendix G)
- Performing off-site evaluation activities (no more than 1 time per 2 year period)
- Analyzing Proficiency Test samples (set of 3 samples, 2 times per year) and reporting Proficiency Test data
- Hosting on-site evaluation, including independent count of a blind slide, (1 time per 2 or 3 year period)
- Performing follow-up action for failed PT results
- Analyzing and reporting Proficiency Tests for each additional method version (set of 3 samples, 2 times per year)

Response Activities/Year/Laboratory = (22.5 application packages + 22.5 off-site activities + 22.5 on-site evaluations + (45 labs*2 PTs) + (4 labs with additional method versions*2 PTs) + 7 PT failures/45 labs = 3.8 Responses/year/lab

Activities for States with Laboratory Certification Programs:

- Reviewing laboratory evaluation application packages (1 time per laboratory per 2 year period)
- Notifying laboratories of certification status and maintaining a list of certified laboratories on State website (1 time per laboratory per 2 year period)
- Conducting and reviewing off-site evaluation activities (1 time per laboratory per 2 year period)
- Conducting on-site evaluations of the laboratories seeking certification (1 time per laboratory per 2 year period)
- Tracking PT results and coordinating follow-up activities for PT results more than 2 standard deviations below the mean value of all participating labs

Response Activities/Year/State = (22.5 application packages + 22.5 off-site activities + 22.5 on-

site evaluations + (45 labs*2 PTs) + (4 labs with additional method versions*2 PTs) + 7 PT failures)/20 states = 8.6 Responses/year/state

5. The Information Collected - Agency Activities, Collection Methodology, and Information Management

5(a) Agency Activities

Agency (EPA Regions and Office of Ground Water and Drinking Water) activities associated with the Lab QA Program consist of the following:

- Assisting State Certification Officers with review of laboratory evaluation application packages
- Assisting State Certification Officers with off-site evaluation activities, including slide evaluation and online analyst evaluation
- Assisting State Certification Officers with on-site evaluation activities, including independent count of blind slides
- Assisting State Certification Officers with interpretation of PT results
- Maintaining a list of links to State websites and/or certified laboratories on EPA's website

5(b) Collection Methodology and Management

Laboratories Seeking State Certification

Laboratories interested in obtaining State certification of their capability to perform analyses using EPA Method 1622, EPA Method 1623 or EPA Method 1623.1 and continuing to demonstrate, proficient and reliable detection and enumeration of *Cryptosporidium* in surface water sources for public water systems may contact State personnel responsible for laboratory certification. Certified laboratories are responsible for notifying the State of losses of key personnel or essential equipment and changes in policies or procedures that directly affect the validity of data or any other change affecting the capability of the laboratory, including change in laboratory location. Participating laboratories are to also demonstrate ongoing capability and method performance by following all applicable method quality control (QC) procedures, analyzing Proficiency Testing (PT) samples (2 times per year), submitting requested data to the State, and participating in periodic evaluations. The Method procedures have been updated to reflect that the minimum recovery for *Cryptosporidium* in ongoing precision and recovery (OPR) samples is now 33 percent (See Appendix J). Laboratories are to document a minimum of 33 percent recovery for OPR samples in an updated QC chart prior to analysis of LT2ESWTR samples at the frequency required in Section 9.7 of EPA method 1622/1623 and Section 9.8 of Method 1623.1. If a laboratory submits PT results more than 2 standard deviations below the mean value of all participating labs, EPA may recommend additional follow-up action to demonstrate that the laboratory's performance remains acceptable. Additional actions may include submission of PT slides, repeat analyses, providing additional QC data, and investigation of problems with reagents and equipment. Repeated failure to demonstrate laboratory capability and acceptable method performance may result in suspension or downgrading of approval status as outlined in 2009 Federal Register Notice (<https://federalregister.gov/a/E9-4009>) (See Appendix D)."

EPA expects that State certification personnel, or their designee, will request a package

with documentation of laboratory personnel status, equipment maintenance, standard operating procedures, training records, and QC charts. EPA expects that the State will evaluate the package for completeness, and would generally follow these steps:

1. The laboratory will send positive staining control and OPR slides for evaluation by State certification officer or their designee (Appendix H).
2. The laboratory will order on-site audit package consisting of two vials containing 50 mL blind samples, 2 vials of artificial matrix, 2 IMS control samples, and 2 analyst evaluation slides. One slide is required per analyst and a back up slide. These will be used in presence of an auditor; therefore, on-site evaluation date should be within product expiration. Each analyst will perform an independent count of one slide. The results and analyst evaluation slides and associated examination forms will be submitted to a State, if requested.
3. The laboratory will schedule an online analyst evaluation of performance for microscopists to demonstrate their ability to identify *Cryptosporidium* oocysts. An example report from the internet analyst evaluation is shown in Appendix I.
4. The State will conduct one 2-day on-site evaluation that will primarily focus on analyst skills and data recording. Laboratory personnel may be asked to prepare a fresh OPR and positive control for review during the on-site evaluation. In addition, laboratory personnel will be asked to use the blind oocyst suspensions ordered in #2 for spiking reagent water and an IMS control in the presence of an auditor, and then complete the analyses within applicable method holding times and send results to the State (Appendix G).
5. The State will send the laboratory a report after the evaluation is complete. The laboratory is then asked to provide written responses to any deficiencies identified in the report. Provided all responses to the deficiencies cited in the report are acceptable, the State will then base its decision for laboratory certification on PT results, quality of the positive control and OPR slide, slide counts, online analyst evaluation, on-site evaluation and recovery values for blind analyses initiated during the on-site evaluation (See Appendices G, H, and I).

5(c) Small Entity Flexibility

The Lab QA Program is a voluntary program; any entity not interested in gaining approval to support LT2ESWTR is not required to participate.

Small businesses are defined as any business that is independently owned and operated and not dominant in its field as defined by the Small Business Administration (SBA) regulations under Section 3 of the Small Business Act.

Small businesses may opt to seek state recognition of laboratory capability to perform *Cryptosporidium* water analyses using only one version of EPA Method 1622/1623 or EPA Method 1623.1, as opposed to being evaluated for multiple versions, and reduce the burden associated with participation in the Lab QA Program.

5(d) Collection Schedule

Laboratory certification is voluntary.

Laboratories that have successfully completed the audit process will not be evaluated more than once per two-year period.

TSC's oversight of LT2ESWTR laboratories during Round 1 represented an exception to the traditional laboratory certification/accreditation approach. At the time, labs and States had limited experience with *Cryptosporidium* monitoring. With the experience gained by laboratories, as well as States, during the first round of monitoring, all parties should be more comfortable with the monitoring and laboratory requirements, thus enabling interested States to take a primary role in lab certification while EPA provides guidance and technical support. The approach for Round 2 creates consistency with the approach for approval/oversight of laboratories for all other regulated analytes and is provided for in the LT2ESWTR rule. A number of States have already expressed an interest in adding *Cryptosporidium* laboratory approval to their programs and have participated in TSC's *Cryptosporidium* Laboratory Certification Officers Training Course in 2010, 2011 and 2013.

6. Estimating the Burden and Cost of the Collection

6(a) Estimating Respondent Burden

Below are summaries of respondent burden hours for this information collection. EPA consulted with six respondents from the community of laboratories that have voluntarily applied for State certification to perform *Cryptosporidium* analyses using EPA Method 1622, 1623, and 1623.1, and 2 respondents from the community of States integrating certification programs to obtain burden hour estimates. For specific burden breakdowns, refer to the *Participating Laboratories Seeking Certification and State Annual Burden* tables in Appendix K. The burden estimates were also addressed in the Federal Register Notice published in September 2013 (See Appendix E).

Based on the first round of LT2ESWTR monitoring, EPA estimates that 45 laboratories will participate in State Certification Programs, of which 15 are private and 30 are state or local government entities. EPA estimates that 20 States will develop programs to certify *Cryptosporidium* laboratories. Twenty-one states have *Cryptosporidium* laboratories within their state boundaries and 19 states responded positively to an industry survey [ASDWA].

Activities for Laboratories Seeking State Certification

Laboratories seeking State certification for analysis of *Cryptosporidium* will complete an application package, including: laboratory documents with resumes for each staff member; standard operating procedures (SOPs); Ongoing Precision and Recovery charts; and Matrix Spike control charts; training records for all analysts/technicians; data package; proficiency testing (PT) results; and a copy of any certificates as documentation of equipment maintenance. Since laboratories will submit the application no more than 1 time per 2 year period, the number of laboratories expected to submit applications were evenly distributed over a 2 year-period to

estimate burden hours and costs per year (e.g., laboratories seeking certification, 45 laboratories/2 years = approximately 22.5 labs/year). Burden hours associated with submitting the completed application package for the laboratories applying for certification are estimated at 293 labor hours per year.

Laboratories seeking certification will also complete off-site evaluation activities including submitting positive staining control and OPR slides, and performing online analyst evaluation. Since laboratories have to complete off-site evaluation activities no more than 1 time per 2 year period, the number of laboratories expected to complete off-site evaluation activities were evenly distributed over a 2 year-period to estimate burden hours per year (e.g., laboratories seeking certification, 45 laboratories/2 years = approximately 22.5 labs/year). Burden hours associated with completing off-site evaluation activities for the laboratories applying for certification are estimated at 486 labor hours per year.

Each laboratory seeking State certification will participate in a 1 to 2 day on-site evaluation. The burden hours associated with this task include time required to attend short briefings by the auditors before and after the audit, demonstrate the techniques for the methods for which they are seeking certification, participate in discussions with the auditors, and respond to any deficiencies noted in the audit report. Laboratories will purchase blind spikes and slides from a qualified vendor and will process samples to demonstrate method performance and perform an independent count of a blind slide. Because laboratories will undergo an on-site evaluation no more than 1 time every 2 years, the number of laboratories expected to be evaluated were evenly distributed over a 2 year period to estimate burden hours per year (e.g., laboratories seeking certification, 45 laboratories/2 years = approximately 22.5 labs/year). Burden hours for all laboratories applying for certification are estimated at 889 labor hours per year (Appendix K).

Laboratories will analyze a set of proficiency test samples, which may include confounding organisms (3 samples per set) 2 times a year for each method version for which they are seeking certification. The burden estimates associated with this task include labor associated with analyzing samples and documenting the data for each proficiency test set. Burden hours for all laboratories seeking certification and analyzing 1 set of proficiency test samples 2 times a year are estimated at 1541 labor hours per year (Appendix K). It is estimated that 4 laboratories seeking State certification will perform more than 1 method version and will perform an additional set of PT samples for each additional method version 2 times a year. Burden hours for laboratories seeking certification and performing an additional set of PT samples for each additional method version are estimated at 137 labor hours per year (Appendix K). EPA estimates that a small subset of laboratories seeking State certification will have to perform follow-up activities based on inadequate QA/QC, failed OPRs, incomplete records, delayed communication to the State or poor PT results. Burden hours for laboratories seeking certification and performing follow-up activities based on inadequate QA/QC, failed OPRs, incomplete records, delayed communication or poor PT results are estimated at 158 labor hours per year (Appendix K).

Activities for States Integrating *Cryptosporidium* Certification Programs

States integrating certification programs for analysis of *Cryptosporidium* will review a laboratory's application package, including: laboratory documents with resumes for each staff member; standard operating procedures (SOPs); Ongoing Precision and Recovery charts; Matrix Spike control charts; training records for all analysts/technicians; data packages, proficiency testing (PT) results; and a copy of any certificates as documentation of equipment maintenance. Since laboratories will submit the application no more than 1 time per 2 year period, the number of laboratories expected to submit applications were evenly distributed over a 2 year-period to estimate burden hours and costs per year (e.g., laboratories seeking certification, 45 laboratories/2 years = approximately 22.5 labs/year). Burden hours associated with all States reviewing completed application packages for the laboratories applying for certification are estimated at 416 labor hours per year (Appendix K).

States integrating certification programs will also review off-site evaluation activities including positive staining control and OPR slides, and online analyst evaluation. Since laboratories have to complete off-site evaluation activities no more than 1 time per 2 year period, the number of laboratories expected to complete off-site evaluation activities were evenly distributed over a 2 year-period to estimate burden hours per year (e.g., laboratories seeking certification, 45 laboratories/2 years = approximately 22.5 labs/year). Burden hours associated with all States reviewing off-site evaluation activities for the laboratories applying for certification are estimated at 315 labor hours per year (Appendix K).

States will conduct a 1 to 2 day on-site evaluation of each laboratory seeking State recognition of laboratory capability under the Lab QA Program. The burden hours associated with this task include time required to travel to and from the laboratory, conduct short briefings before and after the audit, observe the techniques (including independent count of a blind slide) for the methods for which the laboratory is seeking certification, and participate in discussions with the laboratory personnel. Because laboratories will undergo an on-site evaluation no more than 1 time every 2 years, the number of laboratories expected to be evaluated were evenly distributed over a 2 year period to estimate burden hours per year (e.g., laboratories seeking certification, 45 laboratories/2 years = approximately 22.5 labs/year). Burden hours for States conducting on-site evaluations for laboratories applying for certification are estimated at 450 labor hours per year (Appendix K).

States integrating *Cryptosporidium* certification programs will track and review proficiency test results (3 samples per set) 2 times a year for each method version for which the laboratory is seeking certification. The burden estimates associated with this task include labor associated with receiving, documenting, tracking and reviewing each laboratory's PT results. Burden hours for States tracking and reviewing 1 set of proficiency test samples 2 times a year are estimated at 135 labor hours per year (Appendix K). It is estimated that 4 laboratories seeking State certification will perform more than 1 method version and will perform an additional set of PT samples for each additional method version 2 times a year. Burden hours for States reviewing an additional set of PT samples for each additional method version are estimated at 12 labor hours per year (Appendix K). EPA estimates that States will perform follow-up activities with a small subset of laboratories seeking certification based on QA/QC deficiencies, failed OPRs, incomplete records, delayed communication to the state or PT results more than 2 standard deviations below the mean value of all participating labs. Burden hours for

States coordinating these follow-up activities are estimated at 21 labor hours per year (Appendix K).

States will notify laboratories of certification status after satisfactory completion of all activities. The burden hours associated with this task include time to produce an audit report noting any deficiencies, develop certificates, document communication with the laboratory personnel, and maintain certified laboratory list on State website. Because laboratories will be notified no more than 1 time every 2 years, the number of laboratories expected to be notified were evenly distributed over a 2 year period to estimate burden hours per year (e.g., laboratories seeking certification, 45 laboratories/2 years = approximately 22.5 labs/year). Burden hours for States conducting the laboratory notifications and maintaining certified laboratory lists are estimated at 619 labor hours per year (Appendix K).

6(b) Estimating Respondent Costs

Below are summaries of respondent burden costs for this information collection. EPA consulted with fewer than nine respondents from the community of laboratories that have voluntarily applied for EPA approval of laboratory capability to perform *Cryptosporidium* analyses using EPA Method 1622, 1623, and 1623.1 and States integrating *Cryptosporidium* Certification Programs to obtain burden cost estimates. Respondent costs associated with analysis of PT samples include labor and O&M costs, which are estimated by the laboratories and include cost of analytical sample and PT set. Respondent costs associated with evaluation activities include labor and O&M costs, which are estimated by the laboratories and include application fees and travel fees for auditors. For specific burden breakdowns, refer to the *Participating Laboratories Seeking Certification* and *State Annual Burden* tables in Appendix K.

EPA estimates that 45 laboratories will seek State Certification and 20 States will integrate *Cryptosporidium* into their existing Certification Programs.

Cost for Laboratories Seeking Certification

Burden costs associated with submitting the completed application package for the laboratories applying for certification are estimated at \$113,056.88 per year. Burden costs associated with this task include \$24,856.88 for labor and \$88,200.00 for Operations and Maintenance (O&M) (Appendix K).

Burden costs associated with completing off-site evaluation activities for the laboratories are estimated at \$56,846.25 per year. Burden costs associated with this task include \$41,096.25 for labor and \$15,750.00 for O&M (Appendix K).

Burden costs for all laboratories participating in the one 2-day on-site evaluation are estimated at \$150,502.50 per year (Appendix K). Burden costs associated with this task include \$77,287.50 for labor and \$73,215.00 for O&M (Appendix K).

Burden costs for all laboratories analyzing 1 set of PT samples 2 times a year are estimated at \$228,127.50 per year. Burden costs associated with this task include \$128,002.50

for labor and \$100,125.00 for O&M (Appendix K). It is estimated that 4 laboratories seeking State certification will perform more than 1 method version and will perform an additional set of PT samples for each additional method version 2 times a year. Burden costs for laboratories performing an additional set of PT samples for each additional method version are estimated at \$20,278.00 per year. Burden costs associated with this task include \$11,378.00 for labor and \$8,900.00 for O&M (Appendix K). A small subset of laboratories seeking certification will have to perform follow-up activities based on inadequate QA/QC, failed OPRs, incomplete records, delayed communication to EPA or poor PT results. Burden costs for laboratories performing follow-up activities based on inadequate QA/QC, failed OPRs, incomplete records, delayed communication or poor PT results are estimated at \$22,047.67 per year. Burden costs associated with this task include \$13,181.00 for labor and \$8,866.67 for O&M (Appendix K).

Cost for States Integrating *Cryptosporidium* Certification Programs

Burden costs associated with reviewing the completed application package for the laboratories applying for certification are estimated at \$45,123.75 per year. Burden costs associated with this task include \$45,123.75 for labor and \$0.00 for Operations and Maintenance (O&M) (Appendix K).

Burden costs associated with conducting and reviewing off-site evaluation activities for the laboratories applying for certification are estimated at \$34,290.00 per year. Burden costs associated with this task include \$34,290.00 for labor and \$0.00 for O&M (Appendix K).

Burden costs for all States conducting 1- or 2-day on-site evaluation are estimated at \$48,735.00 per year (Appendix K). Burden costs associated with this task include \$48,735.00 for labor and \$0.00 for O&M (Appendix K).

Burden costs for all States reviewing 1 set of PT samples 2 times a year are estimated at \$14,445.00 per year. Burden costs associated with this task include \$14,445.00 for labor and \$0.00 for O&M (Appendix K). It is estimated that States will review results from 4 laboratories seeking certification that perform more than 1 method version and will review an additional set of PT samples for each additional method version 2 times a year. Burden costs for States reviewing an additional set of PT samples for each additional method version are estimated at \$1,284.00 per year. Burden costs associated with this task include \$1,284.00 for labor and \$0.00 for O&M (Appendix K). States will conduct follow-up activities with a small subset of laboratories seeking certification based on inadequate QA/QC, failed OPRs, incomplete records, delayed communication or PT results more than 2 standard deviations below the mean value of all participating labs. Burden costs for States conducting the coordination based on inadequate QA/QC, failed OPRs, incomplete records, delayed communication or poor PT results are estimated at \$2,247.00 per year. Burden costs associated with this task include \$2,247.00 for labor and \$0.00 for O&M (Appendix K).

Burden costs for all States notifying laboratories of certification status after satisfactory completion of all activities and maintaining a list of certified laboratories on state website are estimated at \$66,791.25 per year. Burden costs associated with this task include \$66,791.25 for labor and \$0.00 for O&M (Appendix K).

6(c) Estimating Agency Burden and Costs

Below are Agency burden hours and associated financial costs pertaining to implementation of the Lab QA Program. For a specific breakdown of burden hours and financial costs, refer to the *Agency Annual Burden* table in Appendix K. Costs and burden hours are broken out based on activities completed by the Agency and supporting contractors. EPA estimates an average hourly cost of \$87.25/hour for Agency program management staff. Based on the published schedule of contractor labor rates for the years covered by this program, the average loaded burden hours and costs for contractor labor were estimated at \$62.60/hour for management staff, and \$43.57/hour for technical staff.

Agency burden is estimated based on the labor hours associated with performing each task for each laboratory seeking State certification. To get the total annual cost, hours and costs are then multiplied by the estimated number of respondents and added to the capital and O&M costs. The burden associated with each information collection task is shown in a separate row of the burden table. EPA estimates that 45 laboratories will participate in the EPA Lab QA Program and 20 States will integrate *Cryptosporidium* Certification Programs. Twenty-one states have *Cryptosporidium* laboratories within their state boundaries and 19 states responded positively to an industry survey [ASDWA].

If needed, the Agency will assist State Certification Officers with review of laboratory evaluation application packages. The Agency burden associated with this review is estimated at 461 labor hours and a cost of \$25,224.75 per year.

If needed, the Agency will assist State Certification Officers with off-site evaluation activities, including internet analyst evaluation, and positive control and ongoing precision and recovery (OPR) slide evaluation. The Agency burden associated with off-site evaluation activities is estimated at 394 labor hours and a cost of \$24,058.78 for labor and \$1,775.00 for O&M per year.

If needed, the Agency will assist State Certification Officers with on-site evaluation activities, including independent counts of blind slides by analysts. The Agency burden associated with on-site evaluation activities is estimated at 416 labor hours and a cost of \$28,178.10 per year.

If needed, the Agency will assist State Certification Officers with interpretation of PT results. The Agency burden associated with this activity is estimated at 506 labor hours and a cost of \$36,030.60 per year. The Agency will assist State Certification Officers with interpretation of PT results for additional method versions. The burden associated with this activity is estimated at 45 labor hours and a cost of \$3,202.72 per year.

The Agency will maintain a list of links to State websites and/or certified laboratories on EPA's website. The Agency burden associated with this technical support is estimated at 130 labor hours and a cost of \$8,475.20 per year.

6(d) Estimating the Respondent Universe and Total Burden and Costs

The affected entities include public and private water testing laboratories, and public administrators of environmental protection programs. EPA estimates that 45 laboratories will seek certification, of which 15 are private and 30 are state or local government entities. EPA estimates that 20 States will integrate *Cryptosporidium* into existing certification programs. The respondent total burden and cost are provided in the *Total Respondent and Agency Burden* tables in Appendix K and are described in greater detail in Sections 6(a) - 6(c).

6(e) Bottom Line Burden Hours and Cost Tables

(i) Respondent Tally

Refer to the burden table in Appendix K titled, *Total Respondent and Agency Burden*, for a specific breakdown of the respondent costs. The Lab QA Program will affect approximately 65 respondents. The laboratory and State respondents will engage in 13 different tasks (refer to Section 4(b)(ii)) involving 5,472 labor hours and costing approximately \$508,718.13 per year for labor (Appendix K). Respondents will invest \$0.00 per year in capital/start-up costs and \$295,056.67 per year in O&M costs (Appendix K).

(ii) Agency Tally

Refer to the burden table in Appendix K titled, *Total Agency and Agency Burden*, for a summary of Agency costs. Six Agency tasks are associated with the Lab QA Program. These tasks will involve approximately 1,953 labor hours annually resulting in a cost of \$123,395.15 per year for labor. The Agency will invest approximately \$0.00 per year in capital/start-up costs and \$1,775.00 per year in O&M costs.

6(f) Reasons for Change in Burden

There is an increase of 629 hours in the total estimated respondent burden compared with the ICR currently approved by OMB. Changes in burden have occurred due to inflation, changes in the respondent universe, re-evaluation of hours for tasks, re-evaluation of O&M costs, and improved demonstration of capability. There is also new burden from states integrating *Cryptosporidium* into their existing certification programs, taking over this responsibility from the EPA.

6(g) Burden Statement

The annual reporting and recordkeeping burden for this collection is estimated to average 84.2 hours annually per respondent (laboratory or State) (the combined total hours per year for laboratories seeking certification and States integrating *Cryptosporidium* certification programs divided by 65 total respondents).

Burden means the total time, effort, or financial resources expended by persons to generate, maintain, retain, or disclose or provide information to or for a Federal agency. This

includes the time needed to review instructions; develop, acquire, install, and utilize technology and systems for the purposes of collecting, validating, and verifying information, processing and maintaining information, and disclosing and providing information; adjust the existing ways to comply with any previously applicable instructions and requirements; train personnel to be able to respond to a collection of information; search data sources; complete and review the collection of information; and transmit or otherwise disclose the information. An agency may not conduct or sponsor, and a person is not required to respond to, a collection of information unless it displays a currently valid OMB control number. The OMB control numbers for EPA's regulations are listed in 40 CFR Part 9 (http://ecfr.gpoaccess.gov/cgi/t/text/text-idx?c=ecfr&tpl=/ecfrbrowse/Title40/40cfr92_main_02.tpl) and 48 CFR Chapter 15 (http://www.access.gpo.gov/nara/cfr/waisidx_00/48cfrv6_00.html).

To comment on the Agency's need for this information, the accuracy of the provided burden estimates, and any suggested methods for minimizing respondent burden, including use of automated collection techniques, EPA has established a public docket for this ICR under Docket ID Number EPA-HQ-OW-2002-0011 which is available for online viewing at www.regulations.gov, or in person viewing at the Water Docket in the EPA Docket Center (EPA/DC), WJC West, Room 3334, 1301 Constitution Avenue, NW, Washington, D.C. The EPA Docket Center Public Reading Room is open from 8:30 a.m. to 4:30 p.m., Monday through Friday, excluding legal holidays. The telephone number for the Reading Room is (202) 566-1744, and the telephone number for the Water Docket is (202) 566-2426. An electronic version of the public docket is available at www.regulations.gov. This site can be used to submit or view public comments, access the index listing of the contents of the public docket, and to access those documents in the public docket that are available electronically. When in the system, select "search," then key in the Docket ID Number identified above. Also, you can send comments to the Office of Information and Regulatory Affairs, oir_submission@omb.eop.gov. Please include the EPA Docket ID Number EPA-HQ-OW-2002-0011 and OMB Control Number 2040-0246 in any correspondence.

APPENDIX A

Federal Register Notice:
Laboratory Quality Assurance Evaluation Program/
Information Collection Request
[Published December 29, 2000]

ENVIRONMENTAL PROTECTION AGENCY

[FRL- 7152-6]

Laboratory Quality Assurance Evaluation Program for Analysis of *Cryptosporidium* under the Safe Drinking Water Act; Agency Information Collection: Proposed Collection; Comment Request

AGENCY: Environmental Protection Agency.

ACTION: Notice; Request for Comment.

SUMMARY: Today's notice invites comment on the U.S. Environmental Protection Agency's (EPA's) proposed Laboratory Quality Assurance Evaluation Program for Analysis of *Cryptosporidium* under the Safe Drinking Water Act (Lab QA Program) (Section I). EPA also plans to submit to the Office of Management and Budget (OMB) for review and approval an Information Collection Request (ICR) associated with information collections under the proposed Lab QA Program (Section II). EPA is requesting comments on specific aspects of the proposed Lab QA Program and the ICR. Finally, EPA solicits comments on its intention to seek an emergency clearance from OMB to begin collecting data from laboratories that are interested in participating in the Lab QA Program prior to OMB's final approval of the ICR.

DATES: The Agency requests comments on today's notice. Comments must be received or post-marked by midnight May 3, 2002. If EPA does not receive adverse comments on or before April 3, 2002 regarding EPA's request for an emergency clearance, the Agency intends to seek a 90-day emergency clearance from OMB to begin collecting data from laboratories that are interested in participating in the Lab QA Program.

ADDRESSES: Please send an original and three copies of your written comments and enclosures (including references) to the W-01-17 Comment Clerk, Water Docket (MC-4101), EPA, 1200 Pennsylvania Avenue, NW, Washington, DC 20460. Due to the uncertainty of mail delivery in the Washington, DC area, in order to ensure that all comments are received please send a separate copy of your comments via electronic mail (e-mail) to Mary Ann Feige, EPA, Office of Ground Water and Drinking Water, feige.maryann@epa.gov, or mail to the attention of Mary Ann Feige, EPA, Technical Support Center, 26 West Martin Luther King Drive (MS-140), Cincinnati, Ohio 45268. Hand deliveries should be delivered to: EPA's Water Docket at 401 M Street, SW, Room EB57, Washington, DC 20460. Please make certain to reference EPA ICR No. 2052.02 and OMB Control No. 2040-0229.

FOR FURTHER INFORMATION: For a copy of the ICR, contact Sharon Gonder at EPA by phone at (202) 564-5256 or by email at gonder.sharon@epa.gov or download off the Internet at <http://www.epa.gov/icr> and refer to EPA ICR No. 2052.02. For technical inquiries, contact Mary Ann Feige, EPA, Office of Ground Water and Drinking Water, Technical Support Center, 26 West Martin Luther King Drive (MS-140), Cincinnati, Ohio 45268, fax number, (513) 569-7191, e-mail address, feige.maryann@epa.gov.

SUPPLEMENTARY INFORMATION:

Submission of comments.

Individuals who want EPA to acknowledge receipt of their comments should enclose a self-addressed, stamped envelope. No facsimiles (faxes) will be accepted. Comments may also be submitted electronically to ow-docket@epamail.epa.gov.

Electronic comments must be submitted as an ASCII, WP5.1, WP6.1 or WP8 file avoiding the use of special characters and form of encryption. Electronic comments must be identified by docket number W-01-17. Comments and data will also be accepted on disks in WP5.1, 6.1, 8 or ASCII file format. Electronic comments on this notice may be filed online at many Federal Depository Libraries.

Availability of docket.

The record for this notice has been established under docket number W-01-17, and includes supporting documentation as well as printed, paper versions of electronic comments. The record is available for inspection from 9 a.m. to 4 p.m., Monday through Friday, excluding legal holidays at the Water Docket, EB 57, EPA Waterside Mall, 401 M Street, SW, Washington, DC 20460. For access to docket materials, please call (202) 260-3027 to schedule an appointment.

Section I: Laboratory Quality Assurance Evaluation Program for Analysis of *Cryptosporidium* under the Safe Drinking Water Act

In September 2000, the Stage 2 Microbial and Disinfection Byproducts Federal Advisory Committee (Committee) signed an Agreement in Principle (Agreement) (65 FR 83015, Dec. 29, 2000) (EPA, 2000) with consensus recommendations for two future drinking water regulations: the Long Term 2 Enhanced Surface Water Treatment Rule (LT2ESWTR) and the Stage 2 Disinfectants and Disinfection Byproducts Rule. The LT2ESWTR is to address risk from microbial pathogens, specifically *Cryptosporidium*, and the Stage 2 DBPR is to address risk from disinfection byproducts. The Committee recommended that the LT2ESWTR require public water systems (PWSs) to monitor their source water for *Cryptosporidium* using EPA Method 1622 or EPA Method 1623. Additional *Cryptosporidium* treatment requirements for PWSs would be based on the source water *Cryptosporidium* levels. EPA intends to take into account the Committee's advice and recommendations embodied in the Agreement when developing the regulations.

To support *Cryptosporidium* monitoring under the LT2ESWTR, the Committee Agreement recommended that "compliance schedules for the LT2ESWTR...be tied to the availability of sufficient analytical capacity at approved laboratories for all large and medium-size affected systems to initiate *Cryptosporidium* and *E.coli* monitoring..."(65 FR 83015, Dec. 29, 2000) (EPA, 2000). Further, the Agreement recommended that *Cryptosporidium* monitoring by large and medium systems begin within six months following rule promulgation. Given the time necessary for EPA to approve a sufficient number of laboratories to assure adequate capacity for LT2ESWTR monitoring, EPA

would need to begin laboratory evaluation prior to promulgation of the rule in order to accommodate such an implementation schedule.

Another factor that warrants initiation of the Lab QA Program prior to promulgation of the LT2ESWTR is grandfathering of monitoring data. The Agreement recommends that systems with “historical” *Cryptosporidium* data that are equivalent to data that would be collected under the LT2ESWTR be afforded the opportunity to use those “historical” (grandfathered) data in lieu of collecting new data under LT2ESWTR. EPA intends to propose such grandfathering provisions in the LT2ESWTR. If EPA indicates that laboratories meet the criteria in the Lab QA Program described today prior to finalizing the LT2ESWTR, systems could develop monitoring data prior to the LT2ESWTR in anticipation of using it as grandfathered data.

EPA’s Office of Ground Water and Drinking Water plans to request from OMB an emergency clearance that would enable expeditious implementation of a voluntary Lab QA Program to support *Cryptosporidium* monitoring under the LT2ESWTR. As such, the Agency could begin to evaluate laboratories that can reliably measure for *Cryptosporidium* using EPA Method 1622 and Method 1623. During the effective period of the emergency clearance, EPA intends to submit to OMB for review and approval a final ICR in order to continue data collection for the Lab QA Program.

As part of today’s notice, EPA is inviting comment on the Lab QA Program. Under the Lab QA Program, EPA would evaluate labs on a case-by-case basis through evaluating their capacity and competency to reliably measure for the occurrence of *Cryptosporidium* in surface water using EPA Method 1622 or EPA Method 1623. The intent of this notice is not to propose establishing the Lab QA Program through a rulemaking. Rather, the criteria described in section I.C. are intended to provide guidance to laboratories that are interested in participating in the Lab QA Program.

EPA has not yet proposed rulemaking on use of such “historical” data nor on the methods themselves under the LT2ESWTR. As noted above, EPA intends to propose allowing systems to use equivalent “historical” data in lieu of collecting new data. EPA anticipates the data generated by labs which meet the evaluation criteria would be very high quality, thus increasing the likelihood that such data would warrant consideration as acceptable “grandfathered” data. However, lab evaluation would not guarantee that data generated will be acceptable as “grandfathered” data, nor would failure to meet evaluation criteria necessarily preclude use of “grandfathered” data. For these reasons, EPA is not establishing the Lab QA Program through rulemaking, but rather as a discretionary and voluntary program under the Safe Drinking Water Act, section 1442 (42 USC 300j-1(a)).

A. What is the purpose of the laboratory quality assurance evaluation program?

The purpose of the Lab QA Program is to identify laboratories that can reliably measure for the occurrence of *Cryptosporidium* in surface water. Existing laboratory certification programs do not include *Cryptosporidium* analysis. This program is designed to assess and confirm the capability of laboratories to perform *Cryptosporidium* analyses. The program will assess whether laboratories meet the recommended personnel and laboratory criteria in today’s notice. This evaluation program is voluntary for laboratories. In the LT2ESWTR, however, EPA intends to require systems to use

approved (or certified) laboratories when conducting *Cryptosporidium* monitoring under the LT2ESWTR.

B. Why has EPA selected Methods 1622 and 1623 as the basis for determining the data quality of laboratories that measure for Cryptosporidium?

EPA Method 1622 and EPA Method 1623 were developed as improved alternatives to the ICR Protozoan Method (EPA, 1996). EPA validated Method 1622 for the determination of *Cryptosporidium* in ambient water in August 1998 and distributed an interlaboratory validated draft method in January 1999. In addition, EPA validated Method 1623 for the simultaneous determination of *Cryptosporidium* (and *Giardia*) in ambient water in February 1999 and distributed a validated draft method in April 1999.

In April 2001, EPA revised and updated Method 1622 (EPA-821-R-01-026) (EPA, 2001a) and Method 1623 (EPA-821-R-01-025) (EPA, 2001b) based on the following: laboratory feedback, the development of equivalent filters and antibodies for use with the methods, and method performance data generated during the ICR Supplemental Surveys (EPA, 2001e). The results of these studies are documented in the Method 1622 interlaboratory validation study report (EPA-821-R-01-027) (EPA, 2001c) and the Method 1623 interlaboratory validation study report (EPA-821-R-01-028) (EPA, 2001d).

C. What criteria should I use to determine if my laboratory should apply?

A laboratory that is interested in participating in the Lab QA Program currently should be operating in accordance with its QA plan (developed by the laboratory) for *Cryptosporidium* analyses. In addition, an interested laboratory should demonstrate its capacity and competency to analyze *Cryptosporidium* using the following recommended criteria:

1. Recommended personnel criteria:

Principal Analyst/Supervisor (1 per laboratory) should have:

- BS/BA in microbiology or closely related field
- A minimum of 1 year of continuous bench experience with *Cryptosporidium* and immunofluorescent assay (IFA) microscopy
- A minimum of six months experience using EPA Method 1622 and/or EPA Method 1623
- A minimum of 100 samples analyzed using EPA Method 1622 and/or EPA Method 1623 (minimum 50 samples if the person was an analyst approved to conduct analysis for the ICR Protozoan Method (EPA, 1996)) for the specific analytical procedure they will be using
- Submit to EPA, along with the application package, resumes detailing the qualifications of the laboratory's proposed principal analyst/supervisor

Other Analysts (no minimum number of analysts per laboratory) should have:

- Two years of college (or equivalent) in microbiology or closely related field
- A minimum of six months of continuous bench experience with *Cryptosporidium* and IFA microscopy
- A minimum of three months experience using EPA Method 1622 and/or EPA Method 1623

- A minimum of 50 samples analyzed using EPA Method 1622 and/or EPA Method 1623 (minimum 25 samples if the person was an analyst approved to conduct analysis for the ICR Protozoan Method) for the specific analytical procedures they will be using
- Submit to EPA, along with the application package, resumes detailing the qualifications of the laboratory's proposed other analysts

Technician(s) (no minimum number of technicians per laboratory) should have:

- Three months experience with the specific parts of the procedure they will be performing
- A minimum of 50 samples analyzed using EPA Method 1622 and/or EPA Method 1623 (minimum 25 samples if the person was an analyst approved to conduct analysis for the ICR Protozoan Method) for the specific analytical procedures they will be using
- Submit to EPA, along with the application package, resumes detailing the qualifications of the laboratory's proposed technician(s)

2. Recommended laboratory criteria:

- Appropriate instrumentation as described in EPA Methods 1622 and 1623 (EPA, 2001a,b)
- Equipment and supplies as described in EPA Methods 1622 and 1623 (EPA 2001a, 2001b)
- Detailed laboratory standard operating procedures for each version of the method that the laboratory will use to conduct the *Cryptosporidium* analyses
- Laboratory should provide a current copy of the table of contents of their laboratory's quality assurance plan for protozoa analyses
- EPA Method 1622 or EPA Method 1623 initial demonstration of capability (IDC) data, which include precision and recovery (IPR) test results and matrix spike/matrix spike duplicate (MS/MSD) test results for *Cryptosporidium*. EPA intends to evaluate the IPR and MS/MSD results against the performance acceptance criteria in the April 2001 version of EPA Method 1622 or EPA Method 1623 (EPA, 2001a, 2001b).

D. How can I obtain an application package?

After the OMB clearance described above, EPA plans to make applications available on EPA's website at www.epa.gov/safewater/cryptolabapproval.html. Completed applications should be sent to: EPA's Laboratory Quality Assurance Evaluation Program Coordinator, c/o Dyncorp I&ET, Inc., 6101 Stevenson Avenue, Alexandria, VA 22304-3540. If a laboratory does not have access to the Internet, the laboratory may contact Dyncorp I&ET, Inc. to request an application package.

E. If I demonstrate my laboratory's capacity and competency according to the the personnel and laboratory criteria, what do I do next?

After the laboratory submits to EPA an application package including supporting documentation, EPA intends to conduct the following steps to complete the process:

1. Upon receipt of a complete package, EPA contacts the laboratory for follow-up information and to schedule participation in the performance testing program.
2. EPA sends initial proficiency testing (IPT) samples to the laboratory (unless the laboratory has already successfully analyzed such samples under EPA's Protozoan PE program). IPT samples packets consist of eight spiked samples shipped to the laboratory within a standard matrix.
3. The laboratory analyzes IPT samples and submits data to EPA.
4. EPA conducts an on-site evaluation and data audit.

5. The laboratory analyzes ongoing proficiency testing (OPT) samples three times per year and submits the data to EPA. OPT sample packets consist of three spiked samples shipped to the laboratory within a standard matrix.

6. EPA contacts laboratories by letter within 60 days of their laboratory on-site evaluation to confirm whether the laboratory has demonstrated its capacity and competency for participation in the program.

F. My laboratory has already submitted initial demonstration of capability (IDC) and initial performance testing (IPT) data as part of the EPA Protozoan Performance Evaluation (PE) Program. Do I have to perform this demonstration testing again?

No. If a laboratory currently participates in the EPA Protozoan PE Program and acceptable IDC and IPT data have already been submitted (for the version of the method that the laboratory will use to conduct *Cryptosporidium* analyses), EPA would not expect the laboratory to repeat IDC and IPT analyses.

Section II: Paperwork Reduction Act

The information collection requirements in this notice have been submitted for approval to the OMB under the Paperwork Reduction Act, 44 U.S.C. 3501 et seq. An ICR document has been prepared by EPA (ICR No. 2052.02) and a copy may be obtained from Susan Auby by mail at Collection Strategies Division; EPA (2822); 1200 Pennsylvania Ave., NW, Washington, DC 20460, by email at auby.susan@epamail.epa.gov, or by calling (202) 260-4901. A copy may also be downloaded off the internet at <http://www.epa.gov/icr>.

Since the EPA would solicit information in application packages, including supporting documentation, analytical data, and other pertinent information from laboratories that are interested in participating in the voluntary Lab QA Program, the Agency is required to submit an ICR to OMB for review and approval. Entities potentially affected by this action include public and private laboratories that wish to be evaluated to determine if they can reliably measure for the occurrence of *Cryptosporidium* in surface waters that are used for drinking water sources using EPA Method 1622 or Method 1623.

The burden estimate for the Lab QA Program information collection includes all the burden hours and costs required for gathering information, and developing and maintaining records associated with the Lab QA Program. The annual public reporting and recordkeeping burden for this collection of information is estimated for a total of 60 respondents and an average 78 hours per response for a total of 4,676 hours at a cost of \$123,650. This estimate assumes that laboratories participating in the Lab QA program have the necessary equipment needed to conduct the analyses. Therefore, there are no start-up costs. The estimated total annual capital costs is \$0.00. The estimated Operation and Maintenance (O&M) costs is \$133,880.

Burden means the total time, effort, or financial resources expended by persons to generate, maintain, retain, or disclose or provide information to or for a Federal agency. This includes the time needed to review instructions; develop, acquire, install, and utilize technology and systems for the purposes of collecting, validating, and verifying information, processing and maintaining information, and disclosing and providing information; adjust the existing ways to comply with any previously applicable instructions and requirements; train personnel to be able to respond to a collection of information; search data sources; complete and review the collection of information; and transmit or otherwise disclose the information.

An Agency may not conduct or sponsor, and a person is not required to respond to a collection of information unless it displays a currently valid OMB control number. The OMB control numbers for EPA's regulations are listed in 40 CFR Part 9 and 48 CFR Chapter 15.

Comments are requested on the Agency's need for this information, the accuracy of the provided burden estimates, and any suggested methods for minimizing respondent burden, including through the use of automated collection techniques. Send comments on the ICR to the Director, Collection Strategies Division; EPA (2822); 1200 Pennsylvania Ave., NW, Washington, DC 20460; and to the Office of Information and Regulatory Affairs, Office of Management and Budget, 725 17th St., N.W., Washington, DC 20503, marked "Attention: Desk Officer for EPA." Include the ICR number in any correspondence. Because OMB is required to make a decision concerning the ICR between 30 and 60 days after March 4, 2002, a comment to OMB is best assured of having its full effect if OMB receives it by April 3, 2002. The final ICR approval notice will respond to any OMB or public comments on the information collection requirements contained in today's notice.

References

EPA. 1996. ICR Microbial Laboratory Manual. Office of Research and Development. EPA/600/R-95/178. April 1996.

EPA. 2000. Stage 2 Microbial and Disinfection Byproducts Federal Advisory Committee Agreement in Principle. Federal Register. Vol. 65, pp. 83015-83024. December 29, 2000.

EPA. 2001a. EPA Method 1622: *Cryptosporidium* in Water by Filtration/IMS/FA. Office of Water. Washington, DC 20460. EPA-821-R-01-026. April 2001.

EPA. 2001b. EPA Method 1623: *Cryptosporidium* and *Giardia* in Water by Filtration/IMS/FA. Office of Water. Washington, DC 20460. EPA-821-R-01-025. April 2001.

EPA. 2001c. Interlaboratory Validation Study Results for *Cryptosporidium* Precision and Recovery for EPA Method 1622. Office of Water. Washington, DC 20460. EPA-821-R-01-027. April 2001.

EPA. 2001d. Interlaboratory Validation Study Results for the Determination of *Cryptosporidium* and *Giardia* Using EPA Method 1623. Office of Water. Washington, DC 20460. EPA-821-R-01-028. April 2001.

EPA. 2001e. Implementation and Results of the Information Collection Rule Supplemental Surveys. Office of Water. Washington, DC 20460. EPA-815-R-01-003. February 2001.

APPENDIX B

Federal Register Notice:
Laboratory Quality Assurance Evaluation Program/
Information Collection Request
[Published March 4, 2002]

ENVIRONMENTAL PROTECTION AGENCY
[FRL-]

Agency Information Collection Activities: Submission for OMB Review; Comment

Request; EPA Laboratory Quality Assurance Evaluation Program for Analysis of *Cryptosporidium* under the Safe Drinking Water Act/ Laboratory approval for the Long Term 2 Enhanced Surface Water Treatment Rule

AGENCY: Environmental Protection Agency (EPA).

ACTION: Notice.

SUMMARY: In compliance with the Paperwork Reduction Act (44 U.S.C. 3501 et seq.), this document announces that the following Information Collection Request (ICR) has been forwarded to the Office of Management and Budget (OMB) for review and approval: EPA Laboratory Quality Assurance Evaluation Program for Analysis of *Cryptosporidium* under the Safe Drinking Water Act, OMB Control Number 2040-0246, expiration date of July 31, 2002. The ICR describes the nature of the information collection and its expected burden and cost; where appropriate, it includes the actual data collection instrument.

DATES: Comments must be submitted on or before [Insert date 30 days after publication in the FEDERAL REGISTER].

ADDRESSES: Send comments, referencing EPA ICR No.2067.02 and OMB Control No.2040-0246, to the following addresses: Susan Auby, U.S. Environmental Protection Agency, Collection Strategies Division (Mail Code 2822), 1200 Pennsylvania Avenue, N.W., Washington, DC 20460; and to Office of Information and Regulatory Affairs, Office of Management and Budget (OMB), Attention: Desk Officer for EPA, 725 17th Street, N.W., Washington, DC 20503.

FOR FURTHER INFORMATION CONTACT: For a copy of the ICR contact Susan Auby at EPA by phone at (202) 260-4901, by E-mail at auby.susan@epamail.epa.gov, or download off the Internet at <http://www.epa.gov/icr> and refer to EPA ICR No. 2067.02, the ICR number has changed from the last notice. All requests should refer to EPA ICR No. 2067.02 and not EPA ICR No. 2052.02. For technical inquiries, contact Mary Ann Feige, EPA, Office of Ground Water and Drinking Water, Technical Support Center, 26 West Martin Luther King Drive (MS-140), Cincinnati, Ohio 45268, fax number, (513) 569-7191, e- mail address, feige.maryann@epa.gov.

SUPPLEMENTARY INFORMATION:

Title: EPA Laboratory Quality Assurance Evaluation Program for Analysis of *Cryptosporidium* under the Safe Drinking Water Act (OMB Control No. 2040-0246 ; EPA ICR No. 2067.01) expiring 7/31/02 . This is a request for extension of a currently approved collection.

Abstract: Section I: Laboratory Quality Assurance Evaluation Program for Analysis of *Cryptosporidium* Under the Safe Drinking Water Act

In September 2000, the Stage 2 Microbial and Disinfection Byproducts Federal Advisory Committee (Committee) signed an Agreement in Principle (Agreement) (65 FR 83015, Dec. 29, 2000) (EPA, 2000) with consensus recommendations for two future drinking water regulations: The Long Term 2 Enhanced Surface Water Treatment Rule (LT2ESWTR) and the Stage 2 Disinfectants and Disinfection Byproducts Rule. The LT2ESWTR is to address risk from microbial pathogens, specifically *Cryptosporidium*, and the Stage 2 DBPR is to address risk from disinfection byproducts. The Committee recommended that the LT2ESWTR require public water systems (PWSs) to monitor their source water for *Cryptosporidium* using EPA Method 1622 or EPA Method 1623. Additional *Cryptosporidium* treatment requirements for PWSs would be

based on the source water *Cryptosporidium* levels. To support *Cryptosporidium* monitoring under the LT2ESWTR, the Committee Agreement recommended that "compliance schedules for the LT2ESWTR * * * be tied to the availability of sufficient analytical capacity at approved laboratories for all large and medium-size affected systems to initiate *Cryptosporidium* and *E.coli* monitoring * * * " (65 FR 83015, Dec. 29, 2000) (EPA, 2000). Further, the Agreement recommended that *Cryptosporidium* monitoring by large and medium systems begin within six months following rule promulgation. Given the time necessary for EPA to approve a sufficient number of laboratories to assure adequate capacity for LT2ESWTR monitoring, EPA would need to begin laboratory evaluation prior to promulgation of the rule in order to accommodate such an implementation schedule. Another factor that warrants initiation of the Lab QA Program prior to promulgation of the LT2ESWTR is grandfathering of monitoring data. The Agreement recommends that systems with "historical" *Cryptosporidium* data that are equivalent to data that would be collected under the LT2ESWTR be afforded the opportunity to use those "historical" (grandfathered) data in lieu of collecting new data under LT2ESWTR. EPA intends to propose such grandfathering provisions in the LT2ESWTR. If EPA indicates that laboratories meet the criteria in the Lab QA Program described today prior to finalizing the LT2ESWTR, systems could develop monitoring data prior to the LT2ESWTR in anticipation of using it as grandfathered data. Under the Lab QA Program, EPA would evaluate labs' capacity and competency to reliably measure for the occurrence of *Cryptosporidium* in surface water using EPA Method 1622 or EPA Method 1623. The intent of this notice is not to propose establishing the Lab QA Program through a rulemaking. Rather, the criteria described in section I.C. are intended to provide guidance to laboratories that are interested in participating in the Lab QA Program. EPA anticipates the data generated by labs which meet the evaluation criteria would be very high quality, thus increasing the likelihood that such data would warrant consideration as acceptable "grandfathered" data. However, lab evaluation would not guarantee that data generated will be acceptable as "grandfathered" data, nor would failure to meet evaluation criteria necessarily preclude use of "grandfathered" data. For these reasons, EPA is not establishing the Lab QA Program through rulemaking, but rather as a discretionary and voluntary program under the Safe Drinking Water Act, section 1442 (42 USC 300j-1(a)).

A. What Is the Purpose of the Laboratory Quality Assurance Evaluation Program?

The purpose of the Lab QA Program is to identify laboratories that can reliably measure for the occurrence of *Cryptosporidium* in surface water. Existing laboratory certification programs do not include *Cryptosporidium* analysis. This program is designed to assess and confirm the capability of laboratories to perform *Cryptosporidium* analyses. The program will assess whether laboratories meet the recommended personnel and laboratory criteria in today's notice. This evaluation program is voluntary for laboratories. In the LT2ESWTR, however, EPA intends to require systems to use approved (or certified) laboratories when conducting *Cryptosporidium* monitoring under the LT2ESWTR.

B. How Can I Obtain an Application Package?

After the OMB clearance described above, EPA plans to make applications available on EPA's website at www.epa.gov/safewater/cryptolabapproval.html. Completed applications should be sent to: EPA's Laboratory Quality Assurance Evaluation Program Coordinator, c/o DynCorp, 6101 Stevenson Avenue, Alexandria, VA 22304-3540. If a laboratory does not have access to the Internet, the laboratory may contact DynCorp to request an application package. Applications may be submitted at any time.

C. If I Demonstrate My Laboratory's Capacity and Competency According to the Personnel and Laboratory Criteria, What Happens Next?

After the laboratory submits to EPA an application package including supporting documentation, EPA intends to conduct the following steps to complete the process:

- 1) Upon receipt of a complete package, EPA contacts the laboratory for follow-up information and to schedule participation in the performance testing program.
- 2) EPA sends initial proficiency testing (IPT) samples to the laboratory. IPT samples packets consist of eight spiked samples shipped to the laboratory within a standard matrix.
- 3) The laboratory analyzes the IPT samples and submits data to EPA. EPA intends to have the laboratory's IPT data meet the IPT criteria of greater than 10% mean recovery and less than 71% relative standard deviation (these criteria were developed based on results from the first six rounds of the EPA PE program). This approach will be used unless unforeseen circumstances merit a reassessment of the approach.
- 4) EPA conducts an on-site evaluation and data audit. Checklist for evaluation and audit is included in ICR.
- 5) The laboratory analyzes ongoing proficiency testing (OPT) samples three times per year and submits the data to EPA. OPT sample packets consist of three spiked samples shipped to the laboratory within a standard matrix. The results of the laboratory's OPT data must meet the OPT criteria which will be calculated for each round of OPT testing using only the data from that round. EPA intends to calculate the lower limit as less than 2 standard deviations from the pooled mean using log it transformed data and intends to calculate the maximum RSD as 2 times the pooled RSD. This approach will be used unless unforeseen circumstances merit a reassessment of the approach.
- 6) EPA contacts laboratories by letter within 60 days of their laboratory on-site evaluation to confirm whether the laboratory has demonstrated its capacity and competency for participation in the program.

An agency may not conduct or sponsor, and a person is not required to respond to, a collection of information unless it displays a currently valid OMB control number. The OMB control numbers for EPA's regulations are listed in 40 CFR part 9 and 48 CFR Chapter 15. The Federal Register document required under 5 CFR 1320.8(d), soliciting comments on this collection of information was published on March 4, 2002 (FR). Three comments were received.

Comments requested further information on the details of the Lab Quality Assurance Program. In response, EPA has added supplementary information to the ICR, including the program application, which includes the self-audit checklist detailing the items that will be evaluated during the on-site evaluation. EPA also has also developed a webpage to provide further information on the program. The website can be accessed at http://www.epa.gov/safewater/lt2/cla_final.html.

Commenters expressed concern that the Lab QA Program does not address the Agency's obligation under the FACA Agreement in Principle to identify adequate laboratory capacity to implement LT2ESWTR. The Lab QA Program does assess laboratory capacity through questions on the application on current and potential laboratory capacity to analyze *Cryptosporidium* samples and the on site evaluations. This information will be compiled as laboratory applications are received, and will be updated during on-site evaluations. The on-site evaluation will allow EPA to validate lab capacity reported to EPA.

Comments were received on the burden estimates. Because laboratories that wish to begin using EPA Methods 1622 and 1623 are required by the methods to purchase the equipment necessary to demonstrate initial acceptable performance, and because this is a method requirement, rather than a program requirement (laboratories can perform the methods without ever participating in the program), the burden estimates assume that no capital costs will be incurred by laboratories participating in the program over and above the costs that would be incurred simply to use the method. Because the program application requires the laboratories applying for approval under the program to submit initial performance data, laboratories that meet these requirements should already have the capacity to perform Methods 1622 or 1623 and therefore will not incur start-up costs.

Commenters wanted to know if training would be available for labs needing help. EPA will provide limited training to laboratories needing assistance with the performance of Methods 1622 and 1623. Information on training will be posted on EPA's website as it becomes available.

Commenters wanted to know the earliest date that acceptable grandfathered data could be generated. EPA is aware of the issues regarding grandfathered data acceptability and will address these issues in the proposed LT2ESWTR. These issues are outside of the scope of this ICR.

Burden Statement: The annual public reporting and record keeping burden for this collection of information is estimated to average 18 hours per response. Burden means the total time, effort, or financial resources expended by persons to generate, maintain, retain, or disclose or provide information to or for a Federal agency. This includes the time needed to review instructions; develop, acquire, install, and utilize technology and systems for the purposes of collecting, validating, and verifying information, processing and maintaining information, and disclosing and providing information; adjust the existing ways to comply with any previously applicable instructions and requirements; train personnel to be able to respond to a collection of information; search data sources; complete and review the collection of information; and transmit or otherwise disclose the information. **Respondents/Affected Entities:** Testing Laboratories **Estimated Number of Respondents:** 60. **Frequency of Response:** 3 times per year. **Estimated Total Annual Hour Burden:** 4347 hours. **Estimated Total Annualized Capital, O&M Cost Burden:** \$123,380.

Send comments on the Agency's need for this information, the accuracy of the provided burden estimates, and any suggested methods for minimizing respondent burden, including through the use of automated collection techniques to the addresses listed above. Please refer to EPA ICR No. 2067.02 and OMB Control No. 2040-0246 in any correspondence.

APPENDIX C

Federal Register Notice:
Laboratory Quality Assurance Evaluation Program/
Information Collection Request
[Published October 26, 2005]

ENVIRONMENTAL PROTECTION AGENCY
[OW-2002-0011, FRL-7988-7]

Agency Information Collection Activities; Submission to OMB for Review and Approval; Comment Request; Laboratory Quality Assurance Evaluation Program for Analysis of Cryptosporidium Under the Safe Drinking Water Act, EPA ICR Number 2067.03, OMB Control Number 2040-0246

AGENCY: Environmental Protection Agency.

ACTION: Notice.

SUMMARY: In compliance with the Paperwork Reduction Act (44 U.S.C. 3501 et seq.), this document announces that an Information Collection Request (ICR) has been forwarded to the Office of Management and Budget (OMB) for review and approval. This is a request to renew an existing approved collection. This ICR is scheduled to expire on October 31, 2005. Under OMB regulations, the Agency may continue to conduct or sponsor the collection of information while this submission is pending at OMB. This ICR describes the nature of the information collection and its estimated burden and cost.

DATES: Additional comments may be submitted on or before November 25, 2005.

ADDRESSES: Submit your comments, referencing docket ID number OW- 2002-0012, to (1) EPA online using EDOCKET (our preferred method), by email to ow-docket@epamail.epa.gov, or by mail to: EPA Docket Center, Environmental Protection Agency, Water Docket, Mail Code 4101T, 1200 Pennsylvania Ave., NW., Washington, DC 20460.

FOR FURTHER INFORMATION CONTACT: Sean Conley, Office of Groundwater and Drinking Water, (Mail Code 4607M), Environmental Protection Agency, 1200 Pennsylvania Ave., NW., Washington, DC 20460; telephone number: 202-564- 1781; fax number: 202-564-3767; e-mail address: conley.sean@epa.gov. For technical inquiries, contact Carrie Moulton, EPA, Office of Ground Water and Drinking Water, Technical Support Center, 26 West Martin Luther King Drive (MS-140), Cincinnati, Ohio 45268; fax number: (513) 569-7191; email address: moulton.carrie@epa.gov.

SUPPLEMENTARY INFORMATION: EPA has submitted the following ICR to OMB for review and approval according to the procedures prescribed in 5 CFR 1320.12. On June 3, 2005 (70 FR 32607), EPA sought comments on this ICR pursuant to 5 CFR 1320.8(d). EPA has addressed the comments received.

EPA has established a public docket for this ICR under Docket ID No. OW- 2002-0012, which is available for public viewing at the Water Docket in the EPA Docket Center (EPA/DC), EPA West, Room B102, 1301 Constitution Ave., NW., Washington, DC. The EPA Docket Center Public Reading Room is open from 8:30 a.m. to 4:30 p.m., Monday through Friday, excluding legal holidays. The telephone number for the Reading Room is (202) 566-1744, and the telephone number for the Water Docket is (202) 566-2426. An electronic version of the public docket is available through EPA Dockets (EDOCKET) at <http://www.epa.gov/edocket>. Use EDOCKET to submit or view public comments, access the index listing of the contents of the public docket, and to access those documents in the public docket that are available electronically. Once in the system, select "search," then key in the docket ID number identified above.

Any comments related to this ICR should be submitted to EPA and OMB within 30 days of this notice. EPA's policy is that public comments, whether submitted electronically or in paper, will be made available for public viewing in EDOCKET as EPA receives them and without change, unless the comment contains copyrighted material, CBI, or other information whose public disclosure is restricted by statute. When EPA identifies a comment containing

copyrighted material, EPA will provide a reference to that material in the version of the comment that is placed in EDOCKET. The entire printed comment, including the copyrighted material, will be available in the public docket. Although identified as an item in the official docket, information claimed as CBI, or whose disclosure is otherwise restricted by statute, is not included in the official public docket, and will not be available for public viewing in EDOCKET. For further information about the electronic docket, see EPA's Federal Register notice describing the electronic docket at 67 FR 38102 (May 31, 2002), or go to <http://www.epa.gov/edocket>.

Title: Laboratory Quality Assurance Evaluation Program for Analysis of Cryptosporidium under the Safe Drinking Water Act.

Abstract: Under the Laboratory Quality Assurance Evaluation Program, EPA evaluates labs on a case-by-case basis through evaluating their capacity and competency to reliably measure for the occurrence of Cryptosporidium in surface water using EPA Method 1622 or EPA Method 1623. To obtain approval under the program, the laboratory must submit an application package and provide: a demonstration of availability of qualified personnel and appropriate instrumentation, equipment and supplies; a detailed laboratory standard operating procedure for each version of the method that the laboratory will use to conduct the Cryptosporidium analyses; a current copy of the table of contents of their laboratory's quality assurance plan for protozoa analyses; and an initial demonstration of capability data for EPA Method 1622 or EPA Method 1623, which include precision and recovery test results and matrix spike/matrix spike duplicate test results for Cryptosporidium. After the laboratory submits to EPA an application package including supporting documentation, EPA and the laboratory conduct the following steps to complete the process:

1. EPA contacts the laboratory for follow-up information and to schedule participation in the performance testing program.

2. EPA sends initial proficiency testing samples to the laboratory (unless the laboratory has already successfully analyzed such samples under EPA's Protozoan PE program). These sample packets consist of eight spiked samples shipped to the laboratory within a standard matrix.

3. The laboratory analyzes initial proficiency testing samples and submits data to EPA.

4. EPA conducts an on-site evaluation and data audit.

5. The laboratory analyzes ongoing proficiency testing samples three times per year and submits the data to EPA. These sample packets consist of three spiked samples shipped to the laboratory within a standard matrix.

6. EPA contacts laboratories by letter within 60 days of their laboratory onsite evaluation to confirm whether the laboratory has demonstrated its capacity and competency for participation in the program.

The procedure for obtaining an application package, the criteria for demonstrating capacity and competency, and other guidance to laboratories that are interested in participating in the program, are provided at http://www.epa.gov/safewater/lt2/cla_final.html.

An agency may not conduct or sponsor, and a person is not required to respond to, a collection of information unless it displays a currently valid OMB control number. The OMB control numbers for EPA's regulations in 40 CFR are listed in 40 CFR part 9 and are identified on the form and/or instrument, if applicable.

Burden Statement: The annual public reporting and recordkeeping burden for this collection of information is estimated to average 19 hours per response. Burden means the total time, effort, or financial resources expended by persons to generate, maintain, retain, or disclose or provide information to or for a Federal agency. This includes the time needed to review instructions; develop, acquire, install, and utilize technology and systems for the purposes of collecting, validating, and verifying information, processing and maintaining information, and disclosing and providing information; adjust the existing ways to comply with any previously

applicable instructions and requirements; train personnel to be able to respond to a collection of information; search data sources; complete and review the collection of information; and transmit or otherwise disclose the information.

Respondents/Affected Entities: Public and private water testing laboratories.

Estimated Number of Respondents: 22.

Frequency of Response: Three times per year.

Estimated Total Annual Hour Burden: 3,980.

Estimated Total Annual Cost: \$275,000, includes \$109,000 annualized capital or O&M costs.

Changes in the Estimates: There is a decrease of 367 hours in the total estimated burden currently identified in the OMB Inventory of Approved ICR Burdens. This decrease is just an adjustment to the estimate.

Dated: October 18, 2005.

Sara Hisel-McCoy,
Acting Director, Collection Strategies Division.

[FR Doc. 05-21370 Filed 10-25-05; 8:45 am] BILLING CODE 6560-50-P

APPENDIX D

Federal Register Notice:
Laboratory Quality Assurance Evaluation Program/
Information Collection Request
[Published February 25, 2009]

ENVIRONMENTAL PROTECTION AGENCY

[EPA-HQ-OW-2002-0011; FRL-8776-6]

Agency Information Collection Activities; Proposed Collection; Comment Request; Laboratory Quality Assurance Evaluation Program for Analysis of *Cryptosporidium* Under the Safe Drinking Water Act (Renewal); EPA ICR No. 2067.04, OMB Control No. 2040-0246

AGENCY: Environmental Protection Agency (EPA).

ACTION: Notice.

SUMMARY: In compliance with the Paperwork Reduction Act (PRA) (44 U.S.C. 3501 et seq.), this document announces that EPA is planning to submit a request to renew an existing approved Information Collection Request (ICR) to the Office of Management and Budget (OMB). This ICR is scheduled to expire on May 31, 2009. This notice describes the current "Laboratory Quality Assurance Evaluation Program for Analysis of *Cryptosporidium* under the Safe Drinking Water Act," hereafter referred to as the "Lab QA Program," and requests comment on both the program and the renewed paperwork requirements.

DATES: Comments must be submitted on or before April 27, 2009.

ADDRESSES: Submit your comments, identified by Docket ID No. EPA-HQ-OW-2002-0011, by one of the following methods:

<http://www.regulations.gov>: Follow the on-line instructions for submitting comments.

Mail: Water Docket, Environmental Protection Agency, Mailcode: 2822T, 1200 Pennsylvania Ave., NW., Washington, DC 20460.

Hand Delivery: Monday through Friday, excluding legal holidays. The telephone number for the Reading Room is (202) 566-1744, and the telephone number for the Water Docket is (202) 566-2426. Such deliveries are only accepted during the Docket's normal hours of operation, and special arrangements should be made for deliveries of boxed information.

Instructions: Direct your comments to Docket ID No. EPA-HQ-OW-2002-0011. EPA's policy is that all comments received will be included in the public docket without change and may be made available online at <http://www.regulations.gov>, including any personal information provided, unless the comment includes information claimed to be Confidential Business Information (CBI) or other information whose disclosure is restricted by statute. Do not submit information that you consider to be CBI or otherwise protected using <http://www.regulations.gov> or e-mail. Please contact EPA prior to submitting CBI. The <http://www.regulations.gov> Web site is an "anonymous access" system, which means EPA will not know your identity or contact information unless you provide it in the body of your comment. If you send an e-mail comment directly to EPA without going through <http://www.regulations.gov> your e-mail address will be automatically captured and included as part of the comment that is placed in the public docket and made available on the Internet. If you submit an electronic comment, EPA recommends that you include your name and other contact information in the body of your comment and with any disk or CD-ROM you submit. If EPA cannot read your comment due to technical difficulties and cannot contact you for clarification, EPA may not be able to consider your comment. Electronic

files should avoid the use of special characters, any form of encryption, and be free of any defects or viruses. For additional information about EPA's public docket visit the EPA Docket Center homepage at <http://www.epa.gov/epahome/dockets.htm>.

FOR FURTHER INFORMATION CONTACT: Carrie Miller, EPA, Office of Ground Water and Drinking Water, Technical Support Center, 26 West Martin Luther King Drive (MS-140), Cincinnati, Ohio 45268; e-mail address: miller.carrie@epa.gov.

SUPPLEMENTARY INFORMATION:

How Can I Access the Docket and/or Submit Comments?

EPA has established a public docket for this ICR under Docket ID No. EPA-HQ-OW-2002-0011, which is available for online viewing at <http://www.regulations.gov>, or in person viewing at the Water Docket in the EPA Docket Center (EPA/DC), EPA West, Room 3334, 1301 Constitution Ave., NW., Washington, DC. The EPA/DC Public Reading Room is open from 8:30 a.m. to 4:30 p.m., Monday through Friday, excluding legal holidays. The telephone number for the Reading Room is 202-566-1744, and the telephone number for the Water Docket is 202-566-2426.

Use <http://www.regulations.gov> to obtain a copy of the draft collection of information, submit or view public comments, access the index listing of the contents of the docket, and to access those documents in the public docket that are available electronically. Once in the system, select "search," then key in the docket ID number identified in this document.

What Information Is EPA Particularly Interested in?

Pursuant to section 3506(c)(2)(A) of the PRA, EPA specifically solicits comments and information to enable it to:

- (i) Evaluate whether the proposed collection of information is necessary for the proper performance of the functions of the Agency, including whether the information will have practical utility;
- (ii) Evaluate the accuracy of the Agency's estimate of the burden of the proposed collection of information, including the validity of the methodology and assumptions used;
- (iii) Enhance the quality, utility, and clarity of the information to be collected; and
- (iv) Minimize the burden of the collection of information on those who are to respond, including through the use of appropriate automated electronic, mechanical, or other technological collection techniques or other forms of information technology, e.g., permitting electronic submission of responses. In particular, EPA is requesting comments from very small businesses (those that employ less than 25) on examples of specific additional efforts that EPA could make to reduce the paperwork burden for very small businesses affected by this collection.

EPA is also interested in any other comments regarding the improvements to the Lab QA Program described in this notice.

What Should I Consider When I Prepare My Comments for EPA?

You may find the following suggestions helpful for preparing your comments:

1. Explain your views as clearly as possible and provide specific examples.

2. Describe any assumptions that you used.
3. Provide copies of any technical information and/or data you used that support your views.
4. If you estimate potential burden or costs, explain how you arrived at the estimate that you provide.
5. Offer alternative ways to improve the collection activity.
6. Make sure to submit your comments by the deadline identified under DATES.
7. To ensure proper receipt by EPA, be sure to identify the docket ID number assigned to this action in the subject line on the first page of your response. You may also provide the name, date, and Federal Register citation.

What Information Collection Activity or ICR Does This Apply to?

Affected entities: Entities potentially affected by this action are public and private water testing laboratories. EPA estimates that a total of 65 laboratories will seek to attain or maintain EPA recognition under the Lab QA Program. This estimate includes 63 laboratories seeking continued recognition under the Lab QA Program and 2 laboratories seeking initial recognition.

Title: Laboratory Quality Assurance Evaluation Program for Analysis of *Cryptosporidium* under the Safe Drinking Water Act (Renewal).

ICR numbers: EPA ICR No. 2067.04, OMB Control No. 2040-0246.

ICR status: This ICR is currently scheduled to expire on May 31, 2009. An Agency may not conduct or sponsor, and a person is not required to respond to, a collection of information, unless it displays a currently valid OMB control number. Approved OMB control numbers for EPA's regulations in title 40 of the CFR are listed in 40 CFR part 9 of the Federal Register and displayed either by publication of the Federal Register or by other appropriate means, such as on the applicable collection instrument or form.

Abstract: In September 2000, the Stage 2 Microbial and Disinfection Byproducts Federal Advisory Committee (Committee) signed an Agreement in Principle (Agreement) (65 FR 83015, December 29, 2000) (EPA, 2000) with consensus recommendations for two future drinking water regulations: the Long Term 2 Enhanced Surface Water Treatment Rule (LT2ESWTR) and the Stage 2 Disinfectants and Disinfection Byproducts Rule. The LT2ESWTR was to address risk from microbial pathogens, specifically *Cryptosporidium*. The Committee recommended that the LT2ESWTR require public water systems (PWSs) to monitor their source water for *Cryptosporidium* using EPA Method 1622 or EPA Method 1623. Additional *Cryptosporidium* treatment requirements for PWSs would be based on the source water *Cryptosporidium* levels. EPA took into account the Committee's advice and recommendations as it developed the LT2ESWTR, which was published on January 5, 2006.

Under the LT2ESWTR, EPA requires public water systems to use approved laboratories when conducting *Cryptosporidium* monitoring. In the preamble to the LT2ESWTR as well as several other notices, EPA has described the criteria for approval of laboratories to analyze *Cryptosporidium* samples under the LT2ESWTR. See 71 FR 727 (January 5, 2006) and 67 FR 9731 (March 4, 2002). The Lab QA Program, as revised, is described in this notice. The purpose of the Lab QA Program is to identify laboratories that can reliably measure for the occurrence of *Cryptosporidium* in surface water and to ensure that approved laboratories maintain that capability. Other, State-based laboratory oversight programs do not currently address approval of laboratories for the *Cryptosporidium* analysis required by the LT2ESWTR.

Through today's notice, EPA is inviting comment on refinements to the information collected to support EPA's Lab QA Program. As of May 2007, EPA concluded that sufficient laboratory capacity exists for the LT2ESWTR. As a result, EPA has generally postponed evaluation of additional laboratories, including commercial, county, municipal and utility laboratories, until further notice. Subject to the availability of resources, EPA will consider evaluation of State and EPA Regional laboratories on a case-by-case basis, based on the role that States and EPA Regions play in the certification and approval programs for laboratories. The Lab QA Program is continuously being refined and updated as new information and technologies become available. The program will continue to evolve and EPA will continue to revise and update burden estimates, as needed, with any subsequent ICR.

Approved laboratories will have demonstrated, and are to continue to demonstrate, proficient and reliable detection and enumeration of *Cryptosporidium* in surface water sources for public water systems. They will have passed all elements in the Lab QA Program and continue to successfully participate in all program activities. Approved laboratories are responsible for notifying EPA of losses of key personnel or essential equipment and changes in policies or procedures that directly affect the validity of data or any other change affecting the capability of the laboratory including change in location. Participating laboratories are to also demonstrate ongoing capability and method performance by following all applicable method quality control (QC) procedures, analyzing ongoing proficiency testing (PT) samples (generally three times per year), submitting requested data to EPA, and participating in periodic re-evaluations.

The Lab QA Program procedures have been updated to reflect that the minimum recovery for *Cryptosporidium* in ongoing precision and recovery (OPR) samples is now 22 percent, updated from the original 11 percent. This updated minimum recovery is based on an updated data set and should provide a better assessment of laboratory performance than the original value for the following reasons: (1) The data set is more current and is based on more samples (a total of 333); (2) 52 more laboratories are included in the data set; (3) data were generated using the 2005 version of Method 1623, which is the required version for LT2ESWTR analyses; (4) data were generated using filters currently used to analyze LT2ESWTR samples rather than those filters used originally; and (5) the number of oocysts spiked into the samples was unknown to the laboratories. Calculations for the updated criteria are available in Docket ID No. EPA-HQ-OW-2002-0011. Laboratories are to now document a minimum of 22 percent recovery for OPR samples in an updated QC chart prior to analysis of LT2ESWTR samples at the frequency required in section 9.7 of the method.

The ongoing PT sample packets generally consist of three spiked samples shipped to the laboratory within a standard matrix. If a laboratory submits poor PT results, EPA may recommend additional follow-up action to demonstrate that the laboratory's performance remains acceptable. Additional actions may include submission of PT slides to EPA, repeat analyses, providing additional QC data, and investigation of problems with reagents and equipment. Repeated failure to demonstrate laboratory capability and acceptable method performance may result in suspension or downgrading of approval status as outlined later in this section.

EPA may re-evaluate laboratories participating in the program to verify *Cryptosporidium* laboratory quality assurance (QA) on both an "as-needed" and periodic basis (generally not exceeding once every three years). In the case of a periodic assessment, EPA will generally notify the laboratory that they are due for re-evaluation and request a package with documentation of personnel status, equipment maintenance, standard operating procedures, training records, and QC charts. After the package has been received, it will be evaluated for

completeness. EPA generally contacts the laboratory within 15 days of package submission if information is missing. When a complete package has been received, the following steps will complete the process:

1. The laboratory will send positive staining control and OPR slides for evaluation by EPA.
2. The laboratory will order blind slides spiked with *Cryptosporidium* from a qualified vendor for each analyst. Each analyst will perform an independent count of one slide. The results and slides will be submitted to a technical auditor.
3. EPA will schedule an on-line Internet analyst verification of performance for microscopists to demonstrate their ability to identify *Cryptosporidium* oocysts.
4. EPA conducts a one-day on-site evaluation that will primarily focus on method performance and data recording. Laboratory personnel will be asked to order blind oocyst suspensions for use in sample and IMS control spiking in the presence of an auditor, and then complete the analyses within applicable method holding times and send results to EPA.
5. EPA will send the laboratory a report detailing all findings, generally within 60 days after the evaluation is complete. The laboratory is then asked to provide written responses to any deficiencies identified in the report within 60 days. Provided all responses to the deficiencies cited in the report are acceptable, the Lab QA Program will then base its decision for continued laboratory approval on PT results, quality of the positive control and OPR slide, slide counts, Internet analyst verification, on-site evaluation and recovery values for blind analyses initiated during the on-site evaluation.

State and EPA Regional Laboratories may contact the laboratory approval manager regarding new application submissions. Subject to available resources, EPA estimates that up to two State or EPA Regional Laboratories will seek first-time approval each year. Laboratories seeking approval under the program must submit an application package and provide: a demonstration of availability of qualified personnel and appropriate instrumentation, equipment and supplies; detailed laboratory standard operating procedures; a current copy of the table of contents of their laboratory's QA plan for protozoa analyses; and an initial demonstration of capability data for EPA Method 1623, which includes initial precision and recovery IPR test results and matrix spike/matrix spike duplicate (MS/MSD) test results for *Cryptosporidium*. After EPA completes its review of the application, the Agency will contact the laboratory for follow-up information and to schedule shipment of initial PT samples consisting of eight spiked samples within a standard matrix. EPA then generally conducts an on-site evaluation and data audit. Further information is provided at http://www.epa.gov/safewater/disinfection/lt2/lab_home.html. The Agency notes that completion of an application by a laboratory does not ensure that the Agency will act on the laboratory's request; interested laboratories are encouraged to contact the laboratory approval manager prior to investing substantial effort towards their application. Further, a decision by the Agency to review an application, to send initial PT samples, and/or to schedule or conduct an on-site evaluation and data evaluation, does not ensure that the review process will be completed or that the laboratory will ultimately be approved. Decisions will be made based on the facts associated with a particular application and actions will be taken as Agency resources permit.

Approved laboratories that do not continue to meet the criteria for the Lab QA Program may have their status downgraded to provisional or have their approval suspended. Details of the basis for downgrading or suspending a laboratory's approval are provided in the section entitled ``Clarification of Basis and Procedures for Downgrading/Suspending Approval for Laboratories for the Analysis of *Cryptosporidium* in Water Under the Long Term 2 Enhanced Surface Water

Treatment Rule" (see the following section). Provided EPA has sufficient resources to review requests for upgrade or reinstatement, laboratories may have to undertake additional activities such as analyzing additional PT samples, undergoing an on-site evaluation, and/or counting blind spiked slides in order to have their status upgraded or their approval reinstated. Details regarding additional activities that may be required are provided in the next section.

Clarification of Basis and Procedures for Downgrading/Suspending Approval of Laboratories for the Analysis of *Cryptosporidium* in Water Under the Long Term 2 Enhanced Surface Water Treatment Rule

EPA's Office of Ground Water and Drinking Water, in the Office of Water, has developed a detailed description of the procedures and criteria used in actions concerning approving, downgrading and suspending laboratories for analysis of drinking water contaminants.

In order to assume primary enforcement responsibility for the drinking water regulations, a State must either have available laboratory facilities, approved by the Administrator, capable of conducting analytical measurements of drinking water contaminants, or establish and maintain its own program for approval of laboratories. States wishing to adapt these procedures and criteria for their own approval program should revise it to accurately reflect their State approval program.

This section is intended to clarify EPA's intended practices and procedures for laboratory approval, downgrading or suspension for analysis of *Cryptosporidium* under the Long Term 2 Enhanced Surface Water Treatment Rule (LT2ESWTR) and to reflect good laboratory practice and standard proficiency evaluation in the industry; it is not a regulation. While EPA intends to generally follow the procedures laid out in this section, not every situation is reflected in these procedures and EPA may need to address case-specific situations in ways that differ from the procedures spelled out here. EPA welcomes comment on these procedures and may decide to revise them at any time in the future to reflect changes to its approach or to clarify and update the text.

``Approved Laboratories" have demonstrated, and continue to demonstrate, proficient and reliable detection and enumeration of *Cryptosporidium* in surface water sources for public water systems. They have passed all elements in the Lab QA Program and continue to successfully participate in all program activities. Approved Laboratories notify the Approval Authority (EPA individual(s) administering the program or State individual(s) administering an equivalent laboratory certification program) of loss of key personnel or essential equipment, change in policies or procedures that directly affect the validity of data, and any other change affecting the capability of the laboratory including change in location.

``Provisionally Approved Laboratories" have deficiencies but demonstrate their ability to consistently produce data of known quality. They continue to successfully participate in all Lab QA Program activities. A Provisionally Approved Laboratory may analyze drinking water samples for LT2ESWTR compliance purposes if the laboratory has identified themselves as provisionally approved to their clients and any reports clearly state that the laboratory's status is ``provisionally approved."

``Not Approved" designates a laboratory that has either not participated in the Lab QA Program, or has applied to the program but possesses deficiencies and, in the opinion of the Approval Authority, does not consistently produce data that has met all applicable method QC requirements or has falsified data.

Basis for Downgrading to "Provisionally Approved" Status

An Approved Laboratory (referred to as "laboratory") may be downgraded to "Provisionally Approved" status for *Cryptosporidium* for any of the following reasons:

Failure to analyze samples for the LT2ESWTR according to the December 2005 version of EPA Method 1623 or EPA Method 1622, including all QA/QC criteria;

Failure to document a minimum of 22 percent for on-going precision and recovery values in an updated QC chart prior to analysis of LT2ESWTR samples at the frequency required in section 9.7 of the method;

Failure to demonstrate proficiency based upon acceptable matrix spike recoveries for all modifications of the method procedures per Section 9.1.2 of the method;

Failure to submit valid Proficiency Test (PT) results or meet PT acceptance limits described by the Approval Authority for the first two initial testing events or two out of three regular testing events administered by a vendor authorized by the Approval Authority. The acceptance limits are laboratory mean recovery between 2 standard deviations (SD) of the mean recovery for all approved laboratories in a given test event. Recoveries below the mean recovery minus 2 SD will fail the PT test event. Recoveries higher than the mean recovery plus 2 SD trigger additional evaluation, which may include one or more of the following: (1) On-site evaluation; (2) presence of a proctor when processing PT samples during the next test event; and/or (3) submission of PT microscope slides to the Approval Authority before the expiration of holding time during the next test event;

Failure to submit PT slides within three weeks of PT test event when requested by the Approval Authority;

Failure to maintain records of method modifications per section 9.1.2.2 of the method;

Failure to notify the Approval Authority of loss of key personnel or essential equipment, change in policies or procedures that directly affect the validity of data, or other changes affecting the capability of the laboratory including change in location. Laboratory Approval does not automatically survive such changes; the Approval Authority may request an on-site or off-site evaluation and/or further proof of compliance with all applicable method requirements;

Failure to submit on-site evaluation materials and any other requested information within the time period requested by the Approval Authority; or

Failure to participate satisfactorily in the Approval Authority Lab QA Program and demonstrate proficiency based upon: Sample and method holding time records; analyst verification skills; relative quality of positive staining control and on-going precision recovery (OPR) slides; acceptable performance of QC checks, including but not limited to blind slide counts; and acceptable precision and recovery values for all method variations.

Procedures for Downgrading to "Provisionally Approved" Status

The Approval Authority will notify the laboratory director or owner of its intent to downgrade after becoming aware of the situation warranting downgrading;

The laboratory director should review the problems cited, and within 30 days of receipt of the letter, send a letter to the Approval Authority specifying immediate corrective actions that are being taken;

The Approval Authority will consider the adequacy of the response and notify the laboratory in writing of its approval status, generally within 14 days of receipt of the laboratory's response;

After the Approval Authority notifies a laboratory, the Approval Authority will post status on the Web site list of laboratories and may schedule an on-site evaluation of the laboratory;

The laboratory should identify and correct its problem(s) to the Approval Authority's satisfaction within 30 days of being notified of the downgrade or have approval status suspended;

A Provisionally Approved laboratory may continue to analyze samples for compliance purposes, but must identify its status as Provisionally Approved on any report;

A laboratory may request that the Approval Authority or State provide technical assistance to help identify and resolve any problem; however, adequate performance is the laboratory's responsibility and Approval Authority assistance should not delay the downgrading procedure.

Basis for Suspending Approval Status

A laboratory may be downgraded from Approved or Provisionally Approved status to "Not Approved" for any of the following reasons:

Repeated verification that all applicable method QC requirements have been followed, when in fact they have not all been met;

Repeated failure to document acceptable OPR values prior to analysis of LT2ESWTR samples;

Reporting PT data from another laboratory as its own;

Falsification of data or other deceptive practices including false verification that data submitted to the Data Collection and Tracking System (DCTS) was generated using approved methods and met all method QA/QC criteria;

Refusal to participate in on-site or off-site evaluations conducted by the Approval Authority.

Basis for Suspending Provisionally Approved Status

Failure to provide a letter to the Approval Authority within 30 days that adequately explains what immediate corrective actions were taken;

Failure to identify and correct problems in response to downgrade within 30 days;

Failure to provide accurate OPR control charts to the Approval Authority;

Failure to submit valid PT results for the next two consecutive authorized PT test events within the acceptance limits specified;

Continued failure to use the analytical methodology specified in the regulations;

Failure to correct deviations identified during an on-site evaluation within 30 days; or

Failure to provide requested demonstration, materials and documentation within 30 days, including: acceptable matrix spike recoveries for all method variations per section 9.1.2 of the method; bench sheets, examination forms or OPR charts for any samples requested; remote analyst verification; recent positive staining control and OPR microscope slides, one of each; and blind slide counts for each analyst.

Procedures for Suspension

The Approval Authority will notify the laboratory, in writing, of its intent to suspend approval. If the laboratory wishes to request reconsideration of this decision, it should submit such a request in writing to the Approval Authority within 30 days of receipt of the notice of intent to suspend approval. The laboratory will generally be downgraded immediately to "provisional approval" in the interim while the suspension is being considered. If no request for reconsideration is filed, approval will be suspended.

The request for reconsideration should be supported with an explanation of the reasons for the challenge and should be signed by a responsible official from the laboratory such as the president/owner for a commercial laboratory, the laboratory supervisor of a municipal laboratory, or the laboratory director for a State or Regional laboratory.

The Approval Authority will make a decision and notify the laboratory in writing, generally within 30 days of receipt of the request for reconsideration. If the request is determined to be valid, the Approval Authority will take appropriate measures to reevaluate the facility and notify the laboratory, in writing, of its decision, generally within 60 days of the reevaluation.

Denial of the request will generally result in suspension of the laboratory's approval. Once approval is suspended, a public water system may not use the laboratory to analyze source water samples for compliance with LT2ESWTR source water monitoring requirements. The laboratory should notify its clients that it is no longer approved and will not accept any more LT2ESWTR samples for analysis.

Upgrading or Reinstatement of Approval

Subject to the availability of resources, the Approval Authority will consider written requests from the laboratory to seek upgrading or reinstatement of approval. Requests should state the reasons why the laboratory should regain its approval status. The laboratory should demonstrate that all deficiencies have been corrected and successfully complete two consecutive authorized PT test events within acceptance limits for Provisionally Approved laboratories or three consecutive authorized PT test events within acceptance limits for suspended laboratories. The authorized PT test events being described here are those submitted to all laboratories in the Lab QA Program, not special issue blind samples purchased independently from the vendor. The laboratory should provide evidence why the reasons for downgrading or suspension are no longer applicable and explain its technical competence. Acceptable demonstration of technical competence may include an on-site evaluation and/or any other measure the Approval Authority deems appropriate. The Approval Authority will consider compliance history, corrective actions implemented by the laboratory, effectiveness of corrective actions, and professional judgment of the Approval Authority.

Grievances

Laboratories with grievances during the authorized PT events or regarding participation in the Lab QA Program should immediately contact the Program Manager at the Approving Authority and try to remedy the problem. When the laboratory feels they have not gotten immediate or satisfactory results, they should contact the supervisor at the Approving Authority. The management at the Approving Authority will work with the Program Manager to quickly

address grievances. A final decision for all grievances will be made generally within 30 days of contacting the Approving Authority.

Request for Comment

The EPA is soliciting comments on this notice to:

1. Evaluate whether the proposed collection of information is necessary for the proper performance of the functions of the Agency, including whether the information will have practical utility;
2. Evaluate the accuracy of the Agency's estimate of the burden of the proposed collection of information, including the validity of the methodology and assumptions used;
3. Enhance the quality, utility, and clarity of the information to be collected;
4. Minimize the burden of the collection of information on those who are to respond, including through use of appropriate automated electronic, mechanical, or other technological collection techniques or other forms of information technology, e.g., permitting electronic submission of responses; and
5. Consider any necessary changes to the Lab QA Program. As an example, EPA is particularly interested in comments from States regarding the potential for their laboratory programs to assume any/all responsibility for the approval and oversight of LT2ESWTR laboratories, including comments on the appropriate timeframes for such. The Agency also welcomes comments regarding the appropriateness of turning to commercial PT providers as the source of PT samples for laboratories, in lieu of the PT program currently administered by the Agency.

Burden Statement: The burden estimate for the Lab QA Program information collection includes all the burden hours and costs required for gathering information, and developing and maintaining records associated with the Lab QA Program. An estimated 65 respondents will participate in an average of 4.4 responses per year to include: analysis and reporting of PT samples three times per year, application for initial or re-audit once every three years, off-site re-evaluation activities once every three years, and on-site evaluation once every three years. A small subset of laboratories will perform follow-up activities based on inadequate QA/QC, failed OPRs, incomplete records, delayed communication to EPA or poor PT results. A few laboratories perform more than one method version and will analyze an additional set of PTs samples three times per year. The total annual public reporting and recordkeeping burden for this collection of information is estimated to be 4843 hours at a cost of \$269,800.40. The average hours and cost per response for the average of 4.4 responses per year are 16.9 hours and \$943.36, respectively. These estimates assume that laboratories participating in the Lab QA Program have the necessary equipment needed to conduct the analyses. Therefore, there are no start-up costs. The estimated total annual capital cost is \$0.00. The total estimated Operation and Maintenance (O&M) costs is \$141,929.00.

The ICR provides a detailed explanation of the Agency's estimate, which is only briefly summarized here:

Estimated Total Number of Potential Respondents: 65.

Frequency of Response: Annual.

Estimated Total Average Number of Responses for Each Respondent: 4.4.

Estimated Total Annual Burden Hours: 4843 hours.

Estimated Total Annual Costs: \$411,729.40. This includes an estimated burden cost of \$269,800.40 and an estimated cost of \$141,929.00 for capital investment or maintenance and operational costs.

Are There Changes in the Estimates From the Last Approval?

Changes in burden have occurred due to inflation, re-evaluation of hours for tasks, and improved demonstration of capability. Inflation has increased all operation and maintenance and labor costs accordingly. The increase in the respondent universe has increased the overall burden costs for the respondents. EPA's original estimates for hours to participate and maintain the Lab QA Program were made before the program began. Because the program has matured and several years of QC data have been collected, the burden has changed for performing improved and refined procedures. The burden for some tasks has been estimated and will be re-evaluated as the program progresses. EPA has added the preceding section entitled ``Clarification of Basis and Procedures for Downgrading/Suspending Approval for Laboratories for the Analysis of *Cryptosporidium* in Water Under the Long Term 2 Enhanced Surface Water Treatment Rule." Some approved laboratories may have to undertake additional activities to demonstrate continued acceptable performance to EPA, which may increase the burden of participation in the Lab QA Program for those laboratories. EPA estimates that nine laboratories per year may have to undertake additional activities to demonstrate acceptable performance to EPA. These estimates will be corrected as the program continues.

What is the Next Step in the Process for This ICR?

EPA will consider the comments received and amend the ICR as appropriate. The final ICR package will then be submitted to OMB for review and approval pursuant to 5 CFR 1320.12. At that time, EPA will issue another Federal Register notice pursuant to 5 CFR 1320.5(a)(1)(iv) to announce the submission of the ICR to OMB and the opportunity to submit additional comments to OMB. If you have any questions about this ICR or the approval process, please contact the technical person listed under FOR FURTHER INFORMATION CONTACT.

Dated: February 19, 2009.
Cynthia C. Dougherty,
Director, Office of Ground Water and Drinking Water.
[FR Doc. E9-4009 Filed 2-24-09; 8:45 am]

BILLING CODE 6560-50-P

APPENDIX E

Federal Register Notice:
Laboratory Quality Assurance Evaluation Program/
Information Collection Request
78 FR 54643, September 5, 2013

[Federal Register Volume 78, Number 172 (Thursday, September 5, 2013)]

[Notices]

[Pages 54643-54644]

From the Federal Register Online via the Government Printing Office [www.gpo.gov]

[FR Doc No: 2013-21637]

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ENVIRONMENTAL PROTECTION AGENCY

[EPA-HQ-OW-2002-0011; FRL-9900-75-OW]

Proposed Information Collection Request; Comment Request; Laboratory Quality Assurance Evaluation Program for Analysis of Cryptosporidium Under the Safe Drinking Water Act (Renewal)

AGENCY: Environmental Protection Agency (EPA).

ACTION: Notice.

SUMMARY: The Environmental Protection Agency is planning to submit an information collection request (ICR), "Laboratory Quality Assurance Evaluation Program for Analysis of Cryptosporidium Under the Safe Drinking Water Act" (EPA ICR No. 2067.05, OMB Control No. 2040-0246) to the Office of Management and Budget (OMB) for review and approval in accordance with the Paperwork Reduction Act (44 U.S.C. 3501 et seq.). Before doing so, EPA is soliciting public comments on specific aspects of the proposed information collection as described below. This is a proposed extension of the ICR, which is currently approved through January 31, 2014. An Agency may not conduct or sponsor and a person is not required to respond to a collection of information unless it displays a currently valid OMB control number.

DATES: Comments must be submitted on or before November 4, 2013.

ADDRESSES: Submit your comments, referencing Docket ID No. EPA-HQ-OW-2002-0011,

online using www.regulations.gov (our preferred method), by email to ow-docket@epa.gov, or by mail to: EPA Docket Center, Environmental Protection Agency, Mail Code 28221T, 1200 Pennsylvania Ave. NW., Washington, DC 20460.

EPA's policy is that all comments received will be included in the public docket without change including any personal information provided, unless the comment includes profanity, threats, information claimed to be Confidential Business Information (CBI) or other information whose disclosure is restricted by statute.

FOR FURTHER INFORMATION CONTACT: Carrie Miller, Technical Support Center (TSC), Office of Ground Water and Drinking Water, (MS-140), Environmental Protection Agency, 26 West Martin Luther King Drive, Cincinnati, Ohio 45268; telephone number: 513-569-7919; fax number:

[[Page 54644]]

513-569-7191; email address: miller.carrie@epa.gov.

SUPPLEMENTARY INFORMATION: Supporting documents which explain in detail the information that the EPA will be collecting are available in the public docket for this ICR. The docket can be viewed online at www.regulations.gov or in person at the EPA Docket Center, EPA West, Room 3334, 1301 Constitution Ave. NW., Washington, DC. The telephone number for the Docket Center is 202-566-1744. For additional information about EPA's public docket, visit <http://www.epa.gov/dockets>.

Pursuant to section 3506(c)(2)(A) of the Paperwork Reduction Act, EPA is soliciting comments and information to enable it to: (i) Evaluate whether the proposed collection of information is necessary for the proper performance of the functions of the Agency, including whether the information will have practical utility; (ii) evaluate the accuracy of the Agency's estimate of the burden of the proposed collection of information, including the validity of the methodology and assumptions used; (iii) enhance the quality, utility, and clarity of the information to be collected; and (iv) minimize the burden of the collection of information on those who are to respond, including through the use of appropriate automated electronic, mechanical, or other technological collection techniques or other forms of information

technology, e.g., permitting electronic submission of responses. EPA will consider the comments received and amend the ICR as appropriate. The final ICR package will then be submitted to OMB for review and approval. At that time, EPA will issue another Federal Register notice to announce the submission of the ICR to OMB and the opportunity to submit additional comments to OMB.

Abstract: Under the Long Term 2 Enhanced Surface Water Treatment Rule (LT2ESWTR), EPA requires public water systems to use approved laboratories when conducting Cryptosporidium monitoring. 40 CFR 141.705(a) provides for approval of Cryptosporidium laboratories by "an equivalent" State laboratory certification program (i.e. equivalent to EPA's Laboratory Quality Assurance Evaluation Program). In the preamble to the LT2ESWTR, as well as several other notices, EPA has described the criteria for approval of laboratories to analyze Cryptosporidium samples under the LT2ESWTR. See 74 FR 8529 (February 25, 2009), 71 FR 727 (January 5, 2006) and 67 FR 9731 (March 4, 2002).

Through today's notice, EPA is inviting comment on refinements to the information collected to support EPA's Lab QA Program. The procedures for Methods 1622, 1623, and 1623.1 (a revision of Method 1623) have been updated to reflect that the minimum recovery for Cryptosporidium in ongoing precision and recovery (OPR) samples is now 33 percent. This minimum recovery is based on an updated data set and should provide a better assessment of laboratory performance than the previous value for the following reasons: (1) The data set is more recent; and (2) the sample size is more than twice as large as the 2009 sample size used to establish the previous value.

State responsibilities for Cryptosporidium laboratory approval and oversight will be comparable to their certification responsibilities for the chemistry and microbiology laboratories that they oversee in their current programs (e.g., initial evaluation of laboratory capability; ongoing assessment of the laboratory--including an assessment of Proficiency Test results; and on-site audits at least triennially). Whereas 40 CFR 142.10(b) generally requires the establishment and maintenance of a laboratory "certification" program for all regulated analytes, State approval programs for Cryptosporidium laboratories are optional based on the structure of the LT2ESWTR (40 CFR 141.705(a)).

If a laboratory is located in a State that does not operate a Cryptosporidium laboratory certification/accreditation program, that laboratory can still support LT2ESWTR monitoring if

the laboratory has been approved by another State's laboratory certification/accreditation program that: (1) Has demonstrated substantial conformity to procedures described in Chapter 7 of ``Supplement 2 to the Fifth Edition of the

Manual for the Certification of Laboratories Analyzing Drinking Water"

<http://water.epa.gov/scitech/drinkingwater/labcert/index.cfm#two> and

(2) uses auditors that have passed the Technical Support Center's (TSC) Cryptosporidium Laboratory Certification Officers Training Course. PWSs should be aware that their States may establish requirements that are more stringent than EPA's regulations; State requirements would take precedence.

Consistent with the longstanding laboratory certification program approach, TSC will: (1) Train State/Regional Certification Officers (CO) responsible for auditing Cryptosporidium laboratories; (2) provide written guidance to State/Regional COs; (3) provide day-to-day technical support to States, Regions, and laboratories; (4) review/assist the Regional programs that oversee State certification/accreditation programs; and (5) maintain a list of links to State Web sites naming certified laboratories and/or a list of certified laboratories on EPA's Web site.

Further information is provided at

http://water.epa.gov/lawsregs/rulesregs/sdwa/lt2/lab_home.cfm.

Form Numbers: None.

Respondents/affected entities: Interested States and Laboratories.

Respondent's obligation to respond: Voluntary.

Estimated number of respondents: 45 labs and 20 States/Territories.

Frequency of response: Annual.

Total estimated burden: 5,472 hours (per year). Burden is defined at 5 CFR 1320.3(b).

Total estimated cost: \$803,774.79 (per year), includes \$295,056.67 annualized capital or operation & maintenance (O&M) costs.

Changes in Estimates: There is an increase of 629 hours in the total estimated respondent burden compared with the ICR currently approved by OMB. Changes in burden have occurred due to inflation, re-evaluation of hours for tasks, re-evaluation of O&M costs, improved demonstration of capability, and integration of laboratory oversight into existing State certification programs (State oversight of laboratories was not addressed in the currently approved burden estimate). The increase in the respondent universe has increased the overall

burden costs for the respondents. As the States implement their certification programs, future estimates will be adjusted.

Dated: August 29, 2013.

Ann Codrington,

Acting Director, Office of Ground Water and Drinking Water.

[FR Doc. 2013-21637 Filed 9-4-13; 8:45 am]

BILLING CODE 6560-50-P

APPENDIX F

Comments on
78 FR 54643, September 5, 2013 Federal Register Notice:
Laboratory Quality Assurance Evaluation Program
Information Collection Request

matt@scientificmethods.com

09/05/2013 12:43PM

To: OW-Docket@EPA

This is in response to Docket ID No. EPA-HQ-OW-2002-0011

It appears that our state is not going to have it's own certification program, so we are depending on the EPA region or an outside agency. We are wondering if there is a date that this will be in place. We have customers already looking for pricing and they are not willing to sign up with us until we are approved for the second round.

We have contacted 3rd partys that could approve us now, but the cost is over \$4500.This will only serve to drive up our costs to our water department customers. So we are relying on the EPA regions for lab approval. We would like to know if EPA is going to require that these individuals be ready to audit and give laboratory approvals by a certain date.

Thank you,

Matt Hayes
Scientific Methods, Inc

American Water Works Association
Government Affairs Office
1300 Eye Street NW
Suite 701W
Washington, DC 20005-3314
T 202.628.8303
F 202.628.2846

November 4, 2013

EPA Docket Center Environmental Protection Agency Mail Code 28221T
1200 Pennsylvania Ave. NW. Washington, DC 20460

RE: Agency Information Collection Activities; Proposals, Submissions, and Approvals:
Laboratory Quality Assurance Evaluation Program for Analysis of *Cryptosporidium*, etc. (EPA-
HQ-OW-2002-0011-0043)

Dear Sir or Madam,

The American Water Works Association (AWWA) appreciates the opportunity to comment on the U.S. Environmental Protection Agency's Information Collection Request, "Laboratory Quality Assurance Evaluation Program for Analysis of *Cryptosporidium* Under the Safe Drinking Water Act" (EPA ICR No. 2067.05, OMB Control No. 2040-0246). Since 1991, the regulation of *Cryptosporidium* in drinking water has occurred through a collaborative partnership between the regulatory community, drinking water suppliers and other stakeholders.

AWWA has been deeply involved throughout this period and was a party to the Microbial Disinfection Byproduct Federal Advisory Committee Agreement in Principle (M/DBP AIP) that underlies the Long-Term 2 Enhanced Surface Water Treatment Rule (LT2ESWTR). AWWA's members familiar with the LT2ESWTR, the ongoing Six-Year Review of the LT2ESWTR, and laboratory practices relevant to this ICR reviewed the proposed ICR. AWWA is committed to the effective implementation of LT2ESWTR and through these comments hopes to improve the LT2ESWTR laboratory approval process so that water systems can collect high quality data during Round 2 monitoring.

April 24, 2012, EPA stated that it would announce if and how it would be revising the LT2ESWTR in December, 2014 (U.S. EPA, *Regulatory Review Process for LT2*, Wynne Miller, EPA Office of Ground Water and Drinking Water Standards and Risk Management Division, April 24, 2012). This date is important because in April 2015, water systems must begin LT2ESWTR Round 2 monitoring. Since the April 2012 meeting EPA has not indicated to water systems whether LT2ESWTR Round 2 monitoring would indeed proceed. Given the lower than anticipated occurrence in Round 1, this is not a small question. At the April 2012 meeting and in related stakeholder meetings, the drinking water community expressed a number of concerns that bear on this ICR and which the Agency should address, including:

- 1) Affirm that Round 2 monitoring is needed and advise water systems that Round 2 monitoring is proceeding as scheduled, or alternatively if it is not proceeding. Notification is needed before December 31, 2013.
- 2) Advise water systems subject to LT2ESWTR on the status and timeline for the current laboratory approval process for Round 2 LT2ESWTR by no later than December 31, 2013 and provide a regularly updated, centralized list of all approved labs and the states that approved each.
- 3) Remove recommendations for use of EPA Method 1623.1 from the authorized activities under this ICR until the method has been robustly tested and the Agency accounts for the method's performance in a revised LT2ESWTR rule as was stipulated in the Microbial/Disinfection Byproduct Agreement in Principle.
- 4) Provide a centralized data management structure to collect Round 2 monitoring data and allow the laboratory approval program to evaluate the ongoing performance of laboratories during Round 2 monitoring (EPA has already discontinued support for the LT2ESWTR data system used to support LT2ESWTR. There is, at present, no plan to fully support LT2ESWTR Round 2 in SDWIS NextGen. See USEPA, SAFE Drinking Water Information System Next Generation, NextGen, Business Requirements Document, BRD, February 13, 2013).
- 5) In order to improve the benefits analysis for any future federal actions, provide states with guidance on how to incorporate genotyping into Round 2 monitoring. Incorporate a mechanism into implementation of Round 2 monitoring that facilitates collecting genotyping data from samples positive for *Cryptosporidium*.
- 6) Correct the draft ICR in the following respects:
 - a) Recognize that EPA has not modified the minimum recovery for *Cryptosporidium* in ongoing precision and recovery (OPR) samples to 33 percent in EPA Methods 1622 and 1623 (78 Federal Register 54644; 74 Federal Register 8529; U.S. EPA, Method 1623: *Cryptosporidium* and *Giardia* in Water by Filtration/IMS/FA, December 2005, EPA 815-R-05-002, as referenced on EPA Laboratory Approval website on October 15, 2013, <http://www.epa.gov/nerlcwww/documents/1623de05.pdf>). The Agency has only included this new minimum recovery in EPA Method 1623.1 (U.S. EPA, Method 1623.1: *Cryptosporidium* and *Giardia* in Water by Filtration/IMS/FA, January 2012, EPA 816-R-12-001, as referenced on EPA Laboratory Approval website on October 15, 2013, <http://water.epa.gov/scitech/drinkingwater/labcert/upload/epa816r12001.pdf>). EPA cannot require use of EPA Method 1623.1 or change the ongoing precision and recovery (OPR) requirements for EPA Methods 1622 or 1623 without a rulemaking, which has yet to occur and for which there is inadequate time prior to laboratory approval for Round 2 monitoring.
 - b) Expand the ICR cost analysis to recognize the shifting of implementation burden for Round 2 laboratory approval from EPA to state agencies, laboratories, and water systems including:

- i) Confusion among regulators and laboratories about how the new system will work.
- ii) Lack of information upon which water systems can organize and budget procurement of laboratory services from appropriately qualified laboratories.
- iii) Increased costs for the laboratory approval process because of (1) the loss of federal subsidy, (2) inefficiencies in the new approach, and (3) additional fee structures imposed by certification bodies.

Each of the above six points directly affect the final ICR and its associated implementation burden. These considerations will dramatically change the burden on states, laboratories, and systems associated with implementing this ICR over what was experienced during Round 1 monitoring.

In addition to the above items, it is important that the Agency and the Office of Management and Budget be aware that additional dialogue with stakeholders is needed on how the Round 2 data will impact treatment requirements at affected facilities. The current rule language recognizes that Round 2 may lead to increasing or reducing treatment expectations. At EPA's stakeholder meetings, staff acknowledged this framework, but it was also clear that additional dialogue is needed to describe how to utilize available data to make an informed treatment selection.

If there is an opportunity to assist EPA to distribute information to drinking water systems so that they can begin procuring laboratory services for Round 2 monitoring, AWWA would be happy to assist the Agency. Similarly, AWWA looks forward to the earliest possible discussion of how Round 2 data should be incorporated into sound treatment selection processes under LT2ESWTR. Please do not hesitate to contact me or Steve Via at, (202) 326-6130 or svia@awwa.org, if you have questions about these comments.

Sincerely,
Thomas W. Curtis Deputy
Executive Director
American Water Works Association

cc: Carrie Miller Wynne Miller Greg Carroll

Who is AWWA?

The American Water Works Association (AWWA) is an international, nonprofit, scientific and educational society dedicated to providing total water solutions assuring the effective management of water. Founded in 1881, the Association is the largest organization of water supply professionals in the world. Our membership includes more than 4,000 utilities that supply roughly 80 percent of the nation's drinking water and treat almost half of the nation's wastewater. Our 50,000-plus total membership represents the full spectrum of the water community: public water and wastewater systems, environmental advocates, scientists, academicians, and others who hold a genuine interest in water, our most important resource.

AWWA unites the diverse water community to advance public health, safety, the economy, and the environment.

APPENDIX G

Example Application for Assessment of *Cryptosporidium* Laboratory QA

Application Package for Assessment of *Cryptosporidium* Laboratory Quality Assurance

Submit electronic package to: [Cert Officer email]
Submit any necessary hard copies to: [Cert Officer Name]
[Address]
[Phone]

Step 1: Submit all requested information

Please submit all requested elements in one package organized as follows. Your application will be evaluated for completeness.

1. Completed Audit Application Form
2. Up to date Standard Operating Procedures (SOPs) for the following:
 - a. Performance of each Method step including: sample spiking, filtration, elution, concentration, purification, slide preparation, sample staining and examination (for each method version, where applicable)
 - b. Reagent preparation
 - c. Cleaning practices
 - d. Corrective action procedures for failing to meet OPR, method blank, staining controls, sample acceptance and analyst verification criteria
 - e. Sampling procedures to be followed by field or utility personnel
 - f. Procedures for data recording, checking manual calculations, and checking accuracy of all data transcriptions
 - g. Procedures for data recording and electronic storage of data, including checking for accuracy of data entry and backup of stored data
3. Training records for all analysts/technicians
4. OPR control chart including at a minimum the last 20 OPR samples processed
5. MS control chart including at a minimum the last 20 MS samples processed
6. Submit two data packages from the last 12 months, include a positive result if available. Include all supporting documentation from the field sample, matrix spike, method blank and positive control slide.
7. NELAP certificate (as applicable)

Step 2: Submit slides for review

An off-site technical auditor will review one recent OPR and associated positive staining control slide. Contact [Cert Officer Name and email] to schedule evaluation of slides. This review may occur prior to or following the on-site audit. The slides prepared for the review during the audit in Step 5 may be requested to fulfill this submission.

Step 3: Order on-site audit package

Order an on-site audit package from Wisconsin State Laboratory of Hygiene (608-224-6260), or equivalent vendor. Each package consists of 2 50 mL blind samples, 2 vials of artificial matrix, 2 IMS control samples, and 2 analyst verification slides. One slide is required per analyst and a back up slide. Additional slides may be added for a fee. These will be used in presence of an auditor; therefore, evaluation date should be within product expiration. Bench sheets and examination forms for the blind samples, and associated method blank and OPR samples should be submitted to [Cert Officer Name and email]. Submit the analyst verification slides and associated examination forms, if requested.

Step 4: Schedule internet analyst evaluation

Contact [Cert Officer Name and email] to schedule internet analyst evaluation for each analyst. A computer with internet connection is needed to complete the session.

Step 5: Prepare fresh OPR and positive staining control for review during the audit

Step 6: Evaluation

The laboratory will receive a report detailing all audit findings. The laboratory should provide complete written responses to any deficiencies or recommendations identified in the report within 60 days. Laboratory status for continued approval will be based on submission of acceptable responses, proficiency test results, the quality of the positive control and OPR slide, slide counts, on-site evaluation, and recovery values for blind samples initiated during audit

***Cryptosporidium* Laboratory Audit Application Form**

Part 1. Laboratory Information

Laboratory Name:		
Address:		
City:	State:	Zip:
Contact Person:		
Title:		
Telephone:	Fax:	
Email address:		
Type of laboratory (check one): <input type="checkbox"/> Commercial <input type="checkbox"/> Utility <input type="checkbox"/> State <input type="checkbox"/> Academic <input type="checkbox"/> Other		
Method used in the Laboratory: <input type="checkbox"/> December 2005 Method 1623 <input type="checkbox"/> January 2012 Method 1623.1		
Number of field samples your laboratory is analyzing per month using Method 1623 and/or 1623.1:		
Date of Previous Audit:	Date of Initial Approval:	

Part 2. Personnel List

Name of Current Analyst and Technicians	Current position (Principal Analyst, Analyst, or Technician)	Evaluated during Previous Audit or Documentation Submitted to EPA (Yes/No)

Part 3. Method and Equipment Information: Versions of Method for which the lab is seeking evaluation

Method Procedure		Key Equipment	Manufacturer/Model
Check all that apply		Provide manufacturer and model for relevant pieces of equipment	
Filtration			
Indicate the volume filtered for each		Cubitainer	
Pall Envirochek® HV		Pump	
IDEXX Filta-Max®		Flow control valve	
Other (describe)		Flow meter or graduated container	
Elution			
Wrist action shaker		Laboratory shaker and side arms	
Filta-Max® wash station		Filta-Max® Manual station	
Other (describe)		Filta-Max® Automatic station	
Concentration			
Centrifugation		Centrifuge - 1500 X G, swinging-bucket centrifuge for 15 mL - 250-mL tubes	
Filtration through membrane			
Other (describe)		Concentrator apparatus (Filta-Max only)	
Purification			
Dynabeads® Crypto		Flat-sided sample tubes	
Dynabeads® CG-combo		Sample mixer/rotator for 10-mL tubes	
Other (describe)		Magnetic particle concentrator for 10-mL tubes	
		Magnetic particle concentrator for 1.5-mL tubes	
Staining and Examination			
Waterborne AquaGlo™		Microscope - Epifluorescence/ differential interference contrast (HMO or DIC) microscope with stage and ocular micrometers	
Waterborne Crypt-a-Glo™			
Waterborne Giardi-a-Glo™		20X to 100X objectives	

Method Procedure		Key Equipment	Manufacturer/Model
Meridian Merifluor®		Excitation/band pass microscope filters for fluorescein isothiocyanate (FITC) assay (provide specifications)	
BTF EasyStain™			
Other (describe)		Excitation/band-pass filters for 4',6-diamidino-2-phenylindole (DAPI) assay (provide specifications)	
Other			
Descriptions of "other" method steps and other comments:		Refrigerator for sample storage	
		Refrigerator for reagent storage	

The above information is complete and accurate to the best of my knowledge.

Name and Signature of Laboratory Director or Designee

Date

Checklist A – Method 1623/1623.1 Audit Package and Data Review

Laboratory Name	Name and Affiliation of Evaluator	Date of Evaluation

Good Laboratory Practice (GLP) is generally defined as a system of management controls for the laboratories to ensure the consistency and reliability of results. Adapted from other federal programs for the purposes of the *Cryptosporidium* Laboratory QA Evaluation Program, GLP includes personnel, equipment, and standard operating procedures appropriate for the program.

Item to be Evaluated	Reference*			Classification	Satisfactory				Comments/ Response Requested
	1623	1623.1	Cert		Yes	No	NA	UNK	
1 Quality Assurance									
1.1 Is documentation (e.g., resume, sample list) available for all Method 1623/1623.1 staff?	9.1	9.1	1.	Requirement GLP					
1.1.1 Have technicians/analysts analyzed the required number of samples using Method 1623 or 1623.1?	22.2	22.2	1.1 -1.3	Requirement GLP					
1.2 Are employee training records available and up to date?	9.1	9.1	1.7	Critical GLP					
1.2.1 Have all analysts documented that they have read and understood the QA Plan and SOPs?	-	-	1.7	Critical GLP					
1.3 Is the laboratory performing analyst verification monthly and does the lab have corrective action procedures in place if criteria are not met?	10.6	9.10	7.1.9	Requirement					
1.3.1 If the laboratory has only one analyst, is the analyst demonstrating analyst verification through comparison with photo libraries or repetitive counts?	10.6.4	9.10.4	7.1.9.5	Recommendation					
1.4 Does the quality assurance plan address requirements for <i>Cryptosporidium</i> analysis under LT2ESWTR?				Critical					
1.5 Have acceptable initial precision and recovery analyses been performed for each version of the method the laboratory is using?	9.1.2.1.1	9.2	7.1.4	Requirement					

Item to be Evaluated	Reference*			Classification	Satisfactory				Comments/ Response Requested
	1623	1623.1	Cert		Yes	No	NA	UNK	
1.6 Of the field/PT samples reviewed, is each field/PT sample associated with an acceptable method blank?	9.6.1	9.7	7.1.2 7.1.5.1	Requirement					
1.6.1 Were all method blanks (MB) evaluated without contamination?	9.6.2.1	9.7.2	7.1.5	Requirement					# MB reviewed:
1.6.2 Were the same lots of reagents (elution, IMS, and staining) used for the method blank and the associated field/PT samples?	-	-	7.1.5.3	Critical					
1.6.3 Is method blank analyzed prior to the analysis of field/PT samples?	9.6	9.7	7.1.5.2	Requirement					
1.7 Is each field/PT sample associated with an acceptable ongoing precision and recovery (OPR) sample?	9.7	9.8	7.1.2 7.1.6.1	Requirement					
1.7.1 What percentage of OPR samples evaluated met the recovery criteria?	9.7.3 Table 3 Table 4	9.8.3 Table 3 Table 4	7.1.6.2						# OPR reviewed:
1.7.2 Were the same lots of reagents (elution, IMS, and staining) used for the OPR and the associated field/PT samples?	-	-	7.1.6.1	Critical					
1.7.3 Is OPR analyzed prior to the analysis of field/PT samples?	9.7.1	9.8.1	7.1.6.2	Requirement					
1.7.4 Does the laboratory maintain control charts of OPR results?	9.7.6 9.1	9.8.3 9.12.1 Table 2	7.1.7	1623 Recommendation 1623.1 Requirement					
1.7.5 What is the mean and relative standard deviation (RSD), or standard deviation, of the recoveries of the OPR samples included in the control chart?	9.4.3 Table 3 Table 4	9.5.3 Table 3 Table 4	7.1.6.2	QC Criteria					Mean: RSD:
1.8 Were matrix spike (MS) samples analyzed at the minimum frequency of 1 MS per 20 (up to and including) field samples from each source?	9.1.8	9.6.1	7.1.2 7.1.10.2	Requirement					# MS reviewed:
1.8.1 Were MS sample volumes within 10% of their associated field samples' volumes?	9.5.1	9.6.2	7.1.10.3	Requirement					
1.8.2 Were MS samples analyzed at the same time and using the same method variation as their associated field samples?	Table 2	Table 2	7.1.10.1	Requirement					

Item to be Evaluated	Reference*			Classification	Satisfactory				Comments/ Response Requested
	1623	1623.1	Cert		Yes	No	NA	UNK	
1.8.3 What is the mean and relative standard deviation of the MS samples reviewed?	Table 3 Table 4 Table 5	Table 3 Table 4 Table 6	-	QC Criteria					Mean: RSD:
1.8.4 Does the laboratory maintain control charts of MS results?	9.5.1.4 9.1	9.6.2.3 9.12.2 Table 2	7.1.10.4	1623 Recommendation 1623.1 Requirement					
1.9 Were OPR samples spiked with 100 - 500 organisms?	9.7	9.8	7.1.6.1	Requirement					
1.10 Does the laboratory perform IMS controls and maintain IMS control charts? If not, how do they troubleshoot low recoveries?	9.7.5.3	9.8.7.3 9.13	-	Recommendation					
1.11 Does the laboratory have an adequate record system for tracking samples, including unique ID, from collection through log-in, analysis, and data reporting?	-	-	8.0	Critical GLP					
1.12 Is the laboratory using the Method December 2005 version of Method 1623 or Method 1623.1 for LT2 samples?				Requirement					
2 Data Recording Procedures									
2.1 Is shipping information complete, i.e., time/date of sample collection, sampler's name, time/date of sample receipt, receiver's initials, sample condition?	8.1.3	8.1.3	8.5	Requirement					
2.1.1 Were all samples evaluated received at ≤20°C and not frozen?	8.1.3	8.1.3	6.3.3	Requirement					
2.2 Do sample numbers on the chain of custody match the sample numbers on the report forms?	-	-	-	Requirement					
2.3 Are current Method 1623/1623.1 bench sheets used to record sample processing data?	-	-	8.2	Recommendation					
2.4 Are all primary measurements during each step recorded, including all raw data used in calculations?	9.1.2.2.5	9.3.5	8.0	Requirement					
2.5 Technician/analyst, date, and time of elution is recorded?	12.2.6.2.1	12.2.7.1 12.3.2.1	8.7	Requirement					
2.6 Technician/analyst, date, and time of slide preparation is recorded?	13.3.3.11	13.3.3.11	8.7	Requirement					

Item to be Evaluated	Reference*			Classification	Satisfactory				Comments/ Response Requested
	1623	1623.1	Cert		Yes	No	NA	UNK	
2.7 Technician/analyst, date, and time of staining is recorded?	14.10	14.10	8.7	Requirement					
2.8 Are batch and lot numbers of reagents used in the analysis of the sample recorded?	-	-	8.7	Critical					
2.8.1 Lot number for the IMS kit is recorded?	-	-	8.7	Critical					
2.8.2 Lot number of the staining kit is recorded?	-	-	8.7	Critical					
2.8.3 Lot number of the spiking suspensions is recorded?	-	-	3.21.4 8.7	Critical					
2.9 Spike value recorded for all spiked samples?	-	-	3.21.4 8.6	Requirement					
2.10 Are Method 1623/1623.1 <i>Cryptosporidium</i> Slide Examination forms used to record sample examination results?	15.2	15.2	8.2	Requirement					
2.11 Name of examining analyst is recorded?	15.2.6	15.2.6	8.7	Requirement					
2.12 Date and time of sample examination is recorded?	15.2.4	15.2.4	8.7	Requirement					
2.13 Are calculations of final concentrations and recoveries complete and correct?	-	-	-	Requirement					
2.14 Is the size of the cysts and oocysts reported to the nearest 0.5 µm?	15.2.2.3 15.2.3.3	15.2.2.4 15.2.3.4	5.4.9.4 5.4.10.4	Requirement					
2.15 Is each reported positive organism detected in a field sample characterized and recorded?	15.2	15.2.2.1 15.2.3.1	5.4.9.1 5.4.10.1	Requirement					
2.16 Do values recorded on the data sheets match the values reported to the client?	-	-	8.1	Requirement					
2.17 Are mistakes on all forms crossed out with a single line, initialed, and dated?	-	-	8.2	Critical					
2.18 Are data always legible and recorded in pen?	-	-	8.2	Critical					
2.19 Was the final report reviewed by QA manager, lab director or an individual other than the analyst?	-	-	8.1	Critical					
2.20 Do records demonstrate each analyst's characterization of 3 oocysts and 3 cysts from positive control for each microscopy session?	15.2.1.1	15.2.1.1	5.4.6	Requirement					
2.21 Data shows that no more than 0.5 mL of pellet was used per IMS?	13.2.4	13.2.3	5.2.3 8.6	Requirement					

Item to be Evaluated	Reference*			Classification	Satisfactory				Comments/ Response Requested
	1623	1623.1	Cert		Yes	No	NA	UNK	
3 Holding Times –Method 1623.1									
3.1	Is sample elution initiated within 96 hours of sample collection or field filtration?	8.2.1 Table 1	8.2.1 Table 5	6.4 8.7	Requirement				
3.2	Are sample elution, concentration, and purification steps completed in one work day?	8.2.2 Table 1	8.2.2 Table 5	6.4 8.7	Requirement				
3.3	Are slides stained within 72 hours of application of the purified sample to the slide?	8.2.3 Table 1	8.2.3 Table 5	6.4 8.7	Requirement				
3.4	Are stained slides read and confirmed within 7 days of staining? [Section 8.2.4 and Table 5]	8.2.4 Table 1	8.2.4 Table 5	6.4 8.7	Requirement				
4 Spike enumeration procedures									
4.1	Source of flow cytometry-enumerated spiking suspensions.	-	11.2	-					
4.2	If 50-L samples are analyzed, what positive control procedure does the laboratory follow for OPR and MS samples: (A) spike entire 50 L, (B) spike and filter 10 L before filtering 40 L, or (C) filter 40 L before spiking and filtering 10 L.	-	-	7.1.10.3					
The following items below are optional if the laboratory is NELAC certified. If the laboratory opts to provide NELAC certification, complete the box below by entering the NELAC certification number and date. Provide copy of certification.									
NELAC Certification Number:					Certification Date:				
5 Laboratory Equipment and Supplies									
5.1 Reagent-grade water testing									
5.1.1	Is reagent water tested monthly for conductivity and total chlorine residual?	-	-	4.3.1	Critical GLP				
5.1.1.1	Were the results for the above parameters acceptable? Total chlorine residual not greater than 0.1 mg/L, conductivity not greater than 2 µmhos/cm?	-	-	4.3.1	Critical GLP				
5.1.2	Has the reagent water been tested annually for metals – Pb, Cd, Cr, Cu, Ni, Zn?	-	-	4.3.1	Critical GLP				

Item to be Evaluated	Reference*			Classification	Satisfactory				Comments/ Response Requested
	1623	1623.1	Cert		Yes	No	NA	UNK	
5.1.2.1 Were the results for the metals testing acceptable; each metal not greater than 0.05 mg/L and collectively not greater than 0.1 mg/L?	-	-	4.3.1	Critical GLP					
5.1.3 Is reagent water tested monthly for heterotrophic plate count?	-	-	4.3.1	Critical GLP					
5.1.3.1 Are the results for the heterotrophic plate count acceptable, < 500 CFU/mL?	-	-	4.3.1	Critical GLP					
5.1.4 Is still or DI unit maintained according to manufacturer's instructions?	-	-	4.3.3	Critical GLP					
5.2 pH meter									
5.2.1 Accuracy ± 0.1 units, scale graduations, 0.1 units?	-	-	3.1.1	Critical GLP					
5.2.2 Is a record maintained for pH measurements and calibrations?	-	-	3.1.4	Critical GLP					
5.2.3 Is pH meter standardized each use period with pH 7, 4 or 10 standard buffers (selection dependent upon desired pH)?	-	-	3.1.4	Critical GLP					
5.2.4 Are all pH buffers dated when received and opened, and discarded before expiration date?	-	-	3.1.5	Critical GLP					
5.3 Balances (top loader or pan balance)									
5.3.1 Are balance calibrations verified monthly using ASTM Class 1, Class 2 or Class 3 weights or weights traceable to Class 1, Class 2, or Class 3 weights, or equivalent? Non-reference weights should be calibrated every six months with reference weights.	-	-	3.2.2	Critical GLP					
5.3.2 Is correction data and Certificate of Traceability available for weights?	-	-	3.2.3	Critical GLP					
5.3.3 Is preventative maintenance conducted yearly at a minimum?	-	-	3.2.4	Recommendation GLP					
5.4 Temperature recording device									

Item to be Evaluated	Reference*			Classification	Satisfactory				Comments/ Response Requested
	1623	1623.1	Cert		Yes	No	NA	UNK	
5.4.1 Are calibration of thermometers checked annually (dial thermometers quarterly) at the temperature used against a reference NIST thermometer or equivalent?	8.1.4	8.1.4	3.3.2	Requirement GLP					
5.4.2 Is the sample storage refrigerator able to maintain temperature of 1 to 10°C?	-	-	3.7.1	Critical GLP					
5.5 Micropipetters									
5.5.1 Have micropipetters been calibrated within the past year?	9.2.1	Appendix A	3.8.2	Requirement GLP					
5.6 Centrifuge									
5.6.1 Is a maintenance contract in place or internal maintenance protocol available?	9.1	9.1	3.15.5	Critical GLP					
5.6.2 Is the centrifuge calibrated yearly?	-	-	3.15.5	Critical GLP					
5.7 Autoclave									
5.7.1 Are date, contents, sterilization time and temperature, and technician initials recorded for each cycle?	-	-	3.5.3	Critical GLP					
5.7.2 Is a maximum registering thermometer or continuous monitoring device used during each autoclave cycle?	-	-	3.5.5	Critical GLP					
5.7.3 Is automatic timing mechanism checked with stopwatch quarterly?	-	-	3.5.6	Critical GLP					
5.7.4 Are spore strips or ampules used monthly to confirm sterilization?	-	-	3.5.5	Critical GLP					
6 Quality Assurance Manual									
6.1 Does the laboratory have a formal QA laboratory plan prepared and ready for examination?	9.1	9.1	4.5.1	Requirement					
6.2 Is a laboratory organization chart or other information available listing staff organization and responsibilities? Does it identify the QA manager and lab director?			4.5.2	Recommendation					
6.2.1 Is the QA manager separate from the lab director?	-	-	4.5.2	Recommendation GLP					

Item to be Evaluated	Reference*			Classification	Satisfactory				Comments/ Response Requested
	1623	1623.1	Cert		Yes	No	NA	UNK	
6.3 Does the laboratory have a schedule and/or procedure for all preventative maintenance of equipment?	-	-	4.5.3	GLP					

Comments:

Checklist B - Laboratory SOP Review

Laboratory Name	Name and Affiliation of Evaluator	Date of Evaluation

Good Laboratory Practice (GLP) is generally defined as a system of management controls for the laboratories to ensure the consistency and reliability of results. Adapted from other federal programs for the purposes of the *Cryptosporidium* Laboratory QA Evaluation Program, GLP includes personnel, equipment, and standard operating procedures appropriate for the program.

Item to be Evaluated For each item, does the SOP specify:	Reference*			Classification	Satisfactory				Comments/ Response Requested
	1623	1623.1	Cert		Yes	No	NA	UNK	
1 Sample Spiking									
1.1 The suspension vial is vortexed for 30 seconds or per manufacturer's instructions?	11.4.3.1.2	11.2.3.2	-	Method Procedure					
1.2 The carboy used for the method blank is randomly selected from carboy stock to check efficacy of cleaning system or disposable carboys are used for all samples?	-	-	7.1.5.3	Critical					
1.3 The details of the suspension vial rinse, including volumes?	11.4.3.1	11.2.3	-	Method Procedure					
1.4 Acceptable sample spiking procedures, including issues not noted in items 1.1 through 1.3?				Critical GLP					
2 Filtration/Elution									
2.1 Envirochek® HV filtration									
2.1.1 The flow rate is maintained at approximately 2 L/min?	12.2.1.2	12.2.1.2	-	Method Procedure					
2.1.2 The volume filtered is measured using a flow totalizer or calibrated carboy?	12.2.4.2	12.2.4.2	-	Requirement					
2.1.3 The sample is stirred during filtration?	12.2.4.1	12.2.4.1	-	Method Procedure					
2.1.4 The details of the carboy rinse after filtration including volume?	12.2.4.5	12.2.4.6	-	Method Procedure					
2.1.5 Appropriate maintenance and cleaning procedures?	-	-	-	Critical					

Item to be Evaluated For each item, does the SOP specify:	Reference*			Classification	Satisfactory				Comments/ Response Requested
	1623	1623.1	Cert		Yes	No	NA	UNK	
2.1.6 Acceptable Envirochek® filtration procedures, including issues not noted in items 2.1.1 through 2.1.5?				Critical GLP					
2.2 Envirochek® HV capsule filter elution									
2.2.1 Measurement of the volume of the elution buffer used or that the volume covers the membrane?	12.2.6.2.2	12.2.8.2	-	Method Procedure					
2.2.2 The speed that samples are shaken?	12.2.6.2.3	12.2.8.3	-	Method Procedure					
2.2.3 The dispersant is added to the sample as per Method 1623.1?		12.2.7	-	1623 Recommendation 1623.1 Requirement					
2.2.4 The samples are shaken three times for 5 minutes each time, and each in a different orientation?	12.2.6.2	12.2.8	-	Method Procedure					
2.2.5 Procedures for filter capsule rinse and addition of rinsate to the centrifuge bottle?	12.2.6.2.8	12.2.8.8	-	Method Procedure					
2.2.6 Acceptable Envirochek® capsule filter elution procedures, including issues not noted in items 2.2.1 through 2.2.5?				Critical GLP					
2.3 Filta-Max® filtration									
2.3.1 The flow rate is maintained at ≤4 L per minute for Filta-Max®?	12.3.1.1.3	12.3.1.1.3	-	Method Procedure					
2.3.2 The volume filtered is measured using a flow totalizer or calibrated carboy?	12.3.1.5.2	12.3.1.5.2	-	Requirement					
2.3.3 Appropriate maintenance and cleaning procedures? [Section 12.3.4]	12.3.4	12.3.4	-	Requirement					
2.3.4 Acceptable Filta-Max® filtration procedures, including issues not noted in items 2.3.1 through 2.3.3?				Critical GLP					
2.4 Filta-Max® filter wash station elution									
2.4.1 The use of PBST to elute the filter?	7.4.2.4	7.6.2.4	-	Method Procedure					
2.4.2 The amount of PBST used for each wash? (approx. 600 mL)	12.3.2.2	12.3.2.2	-	Method Procedure					

Item to be Evaluated For each item, does the SOP specify:	Reference*			Classification	Satisfactory				Comments/ Response Requested
	1623	1623.1	Cert		Yes	No	NA	UNK	
2.4.3 The plunger is moved up and down 20 times during the first wash?	12.3.2.2.1 h	12.3.2.2.1 h	-	Method Procedure					
2.4.4 The plunger is moved up and down gently to avoid generating excess foam?	12.3.2.2.1 h	12.3.2.2.1 h	-	Method Procedure					
2.4.5 That during the second wash the plunger is moved up and down 10 times?	12.3.2.2.2 b	12.3.2.2.2 b	-	Method Procedure					
2.4.6 The instructions for cleaning the wash station between samples?	12.3.4.2	12.3.4.2	-	Requirement					
2.4.7 The housing is rinsed after filter is removed and the rinse is included in the sample volume?	12.3.2.2.1 d	12.3.2.2.1 d	-	Method Procedure					
2.4.8 Acceptable Filta-Max® filter wash station elution procedures, including issues not noted in items 2.4.1 through 2.4.7?				Critical GLP					
3 Concentration									
3.1 Filta-Max® filter sample concentration (as an alternative or in addition to Section 3.2)									
3.1.1 The force of the vacuum is maintained below 30 cm Hg?	NOTE pg 43	NOTE pg 34	-	Method Procedure					
3.1.2 That concentration is performed after each of the washes?	12.3.2.2.1 j	12.3.2.2.1 j	-	Method Procedure					
3.1.3 The sample is concentrated so that some liquid remains above the filter (enough to cover the stir bar about half-way)?	12.3.3.2.1 c	12.3.3.2.1 b	-	Method Procedure					
3.1.4 The stir bar and concentration tube are rinsed after each concentration and the liquid added to the concentrate?	12.3.3.2	12.3.3.2	-	Requirement					
3.1.5 The filter membrane is washed twice with 5 mL of PBST each time?	12.3.3.2.3	12.3.3.2.3	-	Method Procedure					
3.1.6 Acceptable Filta-Max® filter sample concentration procedures, including issues not noted in items 3.1.1 through 3.1.5?				Critical GLP					
3.2 Envirochek® HV and Filta-Max® filter sample centrifugation									

Item to be Evaluated For each item, does the SOP specify:	Reference*			Classification	Satisfactory				Comments/ Response Requested
	1623	1623.1	Cert		Yes	No	NA	UNK	
3.2.1 The sample is centrifuged at 1500 x G (maximum 2000 x G) using a swinging bucket rotor?	13.2.1 including NOTE	13.2.1 including NOTE	-	Method Procedure					
3.2.2 Instructions to ensure the centrifuge tubes are properly balanced prior to centrifugation?	-	-	3.15.4	Critical					
3.2.3 The sample is centrifuged for 15 minutes with start time beginning when centrifuge reaches the required speed?	13.2.1	13.2.1	-	Method Procedure					
3.2.4 The centrifuge is slowly decelerated at the end without using the brake?	13.2.1	13.2.1	-	Method Procedure					
3.2.5 Acceptable Envirochek® HV and Filta-Max® filter sample centrifugation procedures, including issues not noted in items 3.2.1 through 3.2.4?				Critical GLP					
4 Purification and Slide Preparation									
4.1 The centrifuged sample supernatant is aspirated no lower than 5 mL of supernatant above every 0.5 mL of the pellet or portion of 0.5 mL pellet?	13.2.2	13.2.2 13.2.3	5.2.2 5.2.3	Requirement					
4.1.1 The type and internal diameter of pipette used for aspiration of supernatant?	-	NOTE pg 37	-	Recommendation					
4.1.2 The rate of aspiration (i.e., mL/ min or pressure of the vacuum)?	-	13.2.2	-	Recommendation					
4.2 The tube is vortexed vigorously until pellet is completely resuspended?	13.2.3	13.2.2.1	-	Method Procedure					
4.3 Appropriate procedures for dividing pellets greater than 0.5 mL into subsamples and the analysis of the subsamples?	13.2.4	13.2.3	-	Critical					
4.4 No more than 0.5 mL of pellet is used per IMS?	13.2.4	13.2.3	5.2.3	Method Procedure					
4.5 The resuspended pellet volume is quantitatively transferred to the flat-sided tube (2 rinses) including the determination of the rinse volumes?	13.3.2.1	13.3.2.1	-	Method Procedure					
4.6 SL-Buffer A is used at room temperature or that it is checked for precipitate before use?	NOTE pg 47	NOTE pg 39	3.17.2	Method Procedure					
4.7 The volume of 10x SL-Buffer A is 1 mL?	13.3.1.2	13.3.1.2	5.2.5	Method Procedure					

Item to be Evaluated For each item, does the SOP specify:	Reference*			Classification	Satisfactory				Comments/ Response Requested
	1623	1623.1	Cert		Yes	No	NA	UNK	
4.8 The volume of 10x SL-Buffer B is 1 mL?	13.3.1.3	13.3.1.3	5.2.5	Method Procedure					
4.9 Instructions for thorough resuspension of IMS beads prior to addition to the flat-sided tube?	13.3.2.2 13.3.2.4	13.3.2.2 13.3.2.4	-	Method Procedure					
4.10 100 µL of <i>Cryptosporidium</i> and <i>Giardia</i> beads are used?	13.3.2.3 13.3.2.5	13.3.2.3 13.3.2.5	5.2.5	Method Procedure					
4.11 The flat-sided tube is rotated at 18 rpm for 1 hour at room temperature?	13.3.2.6	13.3.2.6	-	Method Procedure					
4.12 Which magnetic concentrators, MPC®-1 or MPC®-6, are used?				Method Procedure					
4.13 The placement of the flat-sided tube in the magnet and the rock technique and time?	13.3.2.9	13.3.2.8 13.3.2.9	-	Method Procedure					
4.14 The sample is quantitatively transferred from the flat-sided tube to the microcentrifuge tube (2 rinses) including rinse volumes?	13.3.2.13	13.3.2.14	-	Method Procedure					
4.15 The flat-sided tube is allowed to sit one minute after each transfer to accumulate residual sample, then the residual is transferred to microcentrifuge tube?	13.3.2.13	13.3.2.14	-	Method Procedure					
4.16 The magnet is in the vertical position in the MPC®-S?	-	13.3.2.13	-	Method Procedure					
4.17 The beads are rinsed with PBS while inside the microcentrifuge tube?	13.3.4	13.3.2.17	-	1623 Recommendation 1623.1 Requirement					
4.18 Standard NaOH (5 µL, 1N) and standard HCl (50 µL, 0.1N) are used?	NOTES pg 49-50	NOTES pg 42	3.17.5	Requirement					
4.19 The sample is vortexed vigorously for 50 seconds immediately after the addition of acid and 30 seconds after the sample has set for 10 minutes at room temperature?	13.3.3	13.3.3	-	Method Procedure					
4.20 The magnet is in the slanted position in the MPC®-S for dissociation steps?	-	13.3.3.6	-	Method Procedure					
4.21 A second dissociation is performed?	13.3.3.10	13.3.3.10	5.2.4	Requirement					

Item to be Evaluated For each item, does the SOP specify:	Reference*			Classification	Satisfactory				Comments/ Response Requested
	1623	1623.1	Cert		Yes	No	NA	UNK	
4.22 When the second dissociation is performed, the laboratory: A) uses a second slide, or B) adds the additional volume to the original slide?	13.3.3.10	13.3.3.10 13.4.5	-	Circle one: A B					
4.23 The volume and the timing of the NaOH addition to the wells?	13.3.3.8	13.3.3.8	-	Method Procedure					
4.24 When the slides are dried (e.g., room temperature or slide warmer), the laboratory: A) uses room temperature, or B) uses 35° to 42°C, or C) follows manufacturer's instructions?	13.3.3.12	13.3.3.12	-	Circle one: A B C					
4.25 If the laboratory has more than one option specified for slide drying, are criteria included for when each option will be used?	-	-	5.3.1	Recommendation					
4.26 That positive and negative staining controls are prepared at the same time the slides are prepared?	14.1	14.1.3	-	Requirement					
4.27 Acceptable sample purification and slide preparation procedures, including issues not noted in items 4.1 through 4.26?				Critical GLP					
5 Sample Staining									
5.1 Which stain to use and to follow manufacturer's instructions for FITC stain application?	14.2	14.2	5.3.2	Method Procedure					
5.2 The slides are incubated in a humid chamber in the dark at room temperature for approximately 30 minutes or per manufacturer's directions?	14.3	14.3	5.3.3	Method Procedure					
5.3 The working DAPI stain is prepared the day it is used?	7.7.2	7.9.2	3.19.2	Method Procedure					
5.4 The stock DAPI is stored at 1 to 10°C in the dark?	7.7.1	7.9.1	3.19.1	Method Procedure					
5.5 The volume of working DAPI applied and the incubation time?	14.6	14.6	-	Method Procedure					
5.6 The technique used to drain the excess stain from the well and to rinse the well?	14.5	14.5	-	Method Procedure					
5.7 What type and amount of mounting media used?	7.8	7.10	-	Method Procedure					

Item to be Evaluated For each item, does the SOP specify:	Reference*			Classification	Satisfactory				Comments/ Response Requested
	1623	1623.1	Cert		Yes	No	NA	UNK	
5.8 That all the edges of the cover slip are sealed well with clear fingernail polish, unless Elvanol® is used?	14.9	14.9	-	Method Procedure					
5.9 The finished slides or slides not read immediately are stored in a humid chamber in the dark at 1° to 10°C (humid chamber not required for Elvanol®)?	14.10	14.10	5.3.6	Method Procedure					
5.10 Acceptable sample staining procedures, including issues not noted in items 5.1 through 5.9?				Critical GLP					
6 Microscope and Examination									
6.1 Instructions for ocular and Kohler adjustments?	10.3.4 10.3.6	10.7 10.8	3.22.10	Requirement					
6.2 That all measurements must be recorded to the nearest 0.5 micron?	15.2.2.3 15.2.3.3	15.2.2.4 15.2.3.4	3.22.5	Requirement					
6.3 Microscope cleaning procedures?	10.4	10.9	3.22.11	Requirement					
6.4 The recording of coordinates of all cysts and oocysts on the worksheet for future reference; and slide orientation on the microscope stage to standardize coordinate recording?	-	-	-	Recommendation					
6.5 The examination and acceptance of positive and negative staining controls before proceeding with examination of field samples?	15.2.1	15.2.1	5.4.6 5.4.7	Requirement					
6.6 That each analyst characterizes 3 oocysts and 3 cysts on the positive staining control at each examination session?	15.2.1.1	15.2.1.1	5.4.6	Requirement					
6.7 Corrective actions if positive and/or negative staining controls are not acceptable?	-	-	5.4.8	Recommendation					
6.8 The criteria for organism identification?	15.2.2	15.2.2 15.2.3	5.4.9 5.4.10	Requirement					
6.9 Every positive organism in a field sample is characterized and recorded?	15.2	15.2.2.1 15.2.3.1	5.4.9.1 5.4.10.1	Requirement					
6.10 Acceptable microscope and examination procedures, including issues not noted in items 6.1 through 6.9?				Requirement GLP					
7 Reagents									

Item to be Evaluated For each item, does the SOP specify:	Reference*			Classification	Satisfactory				Comments/ Response Requested
	1623	1623.1	Cert		Yes	No	NA	UNK	
7.1 Procedures for the preparation of all essential chemicals and reagents?	7.0	7.0	4.2	Critical					
7.2 That expiration dates are specified for all reagents prepared by the laboratory?	-	-	4.2.2	Critical					
8 Quality Assurance									
8.1 Training protocol for new employees?	9.1	9.1	1.7	Requirement GLP					
8.2 Procedures for performing analyst verification?	10.6	9.10	7.1.9	Requirement GLP					
8.3 Positive and interfering organisms detected in field samples are documented by photography?	-	-	5.4.11	Recommendation					
8.4 Acceptable procedures for sample collection for field or utility personnel?	-	-	6.1	Critical GLP					
8.5 Criteria for sample acceptance and corrective action procedures?	8.1.3	8.1.3	6.	Requirement GLP					
8.6 Method required holding times?	8.2	8.2	6.4	Requirement GLP					
8.7 Manual data recording procedures?	-	-	8.0	Critical GLP					
8.8 Procedures for checking the accuracy of data transcriptions, including electronic data entry?	-	-	8.1	Critical GLP					
8.9 Procedures for checking the accuracy of manual calculations?	-	-	8.1	Critical GLP					
8.10 Procedures for electronic data entry and storage?	-	-	8.2	Critical GLP					
8.11 How backup of stored data is performed?	-	-	8.2	Critical GLP					
8.12 Corrective action procedures for OPR failures?	9.7.4	9.8.5	7.1.6.2	Requirement GLP					
8.13 Corrective action procedures for method blank contamination?	9.6.2	9.7.3	7.1.5.2	Requirement GLP					

Item to be Evaluated For each item, does the SOP specify:	Reference*			Classification	Satisfactory				Comments/ Response Requested
	1623	1623.1	Cert		Yes	No	NA	UNK	
8.14 Procedures for identifying and assessing declining trends in recovery through review of control charts and/or other recovery data?	-	-	7.1.7.2	Recommendation GLP					
8.15 Corrective action procedures for investigating QC failures or declining trends in recovery?	-	-	7.1.7.2	Recommendation GLP					
8.16 Acceptable glassware washing procedures?	-	-	4.4	Critical GLP					

Comments:

Checklist C – Method 1623/1623.1 Technical Review – Sample Processing and Microscopy

Laboratory Name	Name and Affiliation of Evaluator	Date of Evaluation

Good Laboratory Practice (GLP) is generally defined as a system of management controls for the laboratories to ensure the consistency and reliability of results. Adapted from other federal programs for the purposes of the *Cryptosporidium* Laboratory QA Evaluation Program, GLP includes personnel, equipment, and standard operating procedures appropriate for the program.

Item to be evaluated	Reference*			Classification	Satisfactory				Comments/ Response Requested
	1623	1623.1	Cert		Yes	No	NA	UNK	
1 Laboratory Facilities									
1.1 Does laboratory appear to have established appropriate safety and health practices prior to use of this method?	5.0	5.0	4.1	Critical					
1.2 Do all laboratory personnel wear gloves when handling biohazard and toxic compounds, and change gloves before touching other surfaces and equipment?	5.3 5.4	5.3	4.1.6	Critical GLP					
1.3 Does the laboratory disinfect bench surfaces before and after analyses?	-	-	4.1.3	Critical GLP					
1.4 Does the laboratory have adequate bench space to perform the method?	-	-	2.0	Critical GLP					
1.5 Other than the issues noted in items 1.1 through 1.4 (if any), no other facility issues were observed?									
2 Reagents									
2.1 Is reagent water used to prepare all reagents?	7.3	7.3	4.3.1	Requirement					
2.2 Are all reagents clearly labeled with identity of reagent, date of preparation, technician initials, and expiration date?	-	-	4.2.2	Critical GLP					

Item to be evaluated	Reference*			Classification	Satisfactory				Comments/ Response Requested
	1623	1623.1	Cert		Yes	No	NA	UNK	
2.3 Are SOPs available in the work area, and does laboratory practice reflect written procedures?				Critical GLP					
3 Sample Spiking				Technician:					
3.1 Was spike suspension vial vortexed for 30 seconds or per manufacturer's instructions?	11.4.3.1.2	11.2.3.2	-	Method Procedure					
3.2 Is the carboy used for method blank randomly selected from carboy stock to check efficacy of cleaning system?	-	-	7.1.5.3	Critical GLP					
3.3 Was the suspension vial adequately rinsed?	11.4.3.1	11.2.3	-	Method Procedure					
3.4 Are SOPs for sample spiking available in the work area, and does laboratory practice reflect written procedures?				Critical GLP					
3.5 Other than issues noted for items 3.1 through 3.4 (if any) was sample spiking demonstrated successfully?									
4 Filtration/Elution									
4.1 Envirochek[®] HV filtration				Technician:					
4.1.1 Are all components required for sample filtration present and in good condition?	6.1 6.2.1-6.2.2 6.3	6.1 - 6.2.8	6.1.7	Requirement GLP					
4.1.2 Is the filter assembly set up correctly?	Figure 3a	Figure 1	-	Method Procedure GLP					
4.1.3 Is the pump adequate for needs?	6.3.3	6.2.4	-	Requirement GLP					
4.1.4 Is the appropriate flow rate maintained (approximately 2 L/min)?	12.2.1.2	12.2.1.2	-	Method Procedure					
4.1.5 Is the volume filtered measured using a flow totalizer or calibrated carboy?	12.2.4.2	12.2.4.2	-	Requirement					

Item to be evaluated	Reference*			Classification	Satisfactory				Comments/ Response Requested
	1623	1623.1	Cert		Yes	No	NA	UNK	
4.1.6 Is the system well maintained and cleaned appropriately following use?	4.5	4.5	6.1.7	Critical GLP					
4.1.7 Is the system able to maintain seal during use with no leaks?	-	-	6.1.7	Requirement GLP					
4.1.8 Are SOPs for Envirochek® HV filtration available in the work area, and does laboratory practice reflect written procedures?				Critical GLP					
4.1.9 Other than issues noted for items 4.1.1 through 4.1.8 (if any) was Envirochek® HV filtration demonstrated successfully?									
4.2 Envirochek® HV capsule filter elution				Technician:					
4.2.1 Is the elution buffer prepared as per Method?	7.4.1	7.6.1	-	Method Procedure					
4.2.2 Is the wrist-shaker assembly set up correctly with arms fully extended?	12.2.6.1.1	12.2.6.1	3.14.2	Method Procedure GLP					
4.2.3 Is the dispersant addition performed as per Method 1623.1?	-	12.2.7	-	1623 Recommendation 1623.1 Method Procedure					
4.2.4 Is volume of elution buffer measured to ensure the use of one 250 mL centrifuge tube?	12.2.6.2.2	12.2.8.2	-	Method Procedure					
4.2.5 Are the samples shaken at an appropriate speed?	12.2.6.2.3	12.2.8.3	3.14.3	Method Procedure					
4.2.6 Are the samples shaken three times for 5 minutes each time, and each in a different orientation?	12.2.6.2	12.2.8	-	Method Procedure					

Item to be evaluated	Reference*			Classification	Satisfactory				Comments/ Response Requested
	1623	1623.1	Cert		Yes	No	NA	UNK	
4.2.7 Are SOPs for Envirochek® HV capsule filter elution available in the work area, and does laboratory practice reflect written procedures?				Critical GLP					
4.2.8 Other than issues noted for items 4.2.1 through 4.2.7 (if any) was Envirochek® HV capsule filter elution demonstrated successfully?									
4.3 Filta-Max® filtration				Technician:					
4.3.1 Which filter is used – Filta-Max® (black end caps) or Filta-Max xpress® (red end caps)?									
4.3.2 Are all components required for sample filtration present and in good condition?	6.1 6.2.1 6.2.3 6.3	6.1 6.2.1-6.2.7 6.2.9	6.1.7	Requirement GLP					
4.3.3 Is the filter assembly set up correctly?	Figure 3b	Figure 2	-	Method Procedure GLP					
4.3.4 Is appropriate flow rate maintained of <4 L per minute for Filta-Max®?	12.3.1.1.3	12.3.1.1.3	-	Method Procedure					
4.3.5 Is the volume filtered measured correctly using a flow meter or calibrated carboy?	12.3.1.5.2	12.3.1.5.2	-	Requirement GLP					
4.3.6 Is system well maintained and cleaned appropriately following use?	12.3.4	12.3.4	6.1.7	Requirement GLP					
4.3.7 Is system able to maintain seal during use with no leaks?	-	-	6.1.7	Requirement GLP					
4.3.8 Does the laboratory indicate on the filter housing the correct direction of flow?	12.3.1.3	12.3.1.3	-	Critical					

Item to be evaluated	Reference*			Classification	Satisfactory				Comments/ Response Requested
	1623	1623.1	Cert		Yes	No	NA	UNK	
4.3.9 Are SOPs for Filta-Max® filtration available in the work area, and does laboratory practice reflect written procedures?				Critical GLP					
4.3.10 Other than issues noted for items 4.3.1 through 4.3.9 (if any) was Filta-Max® filtration demonstrated successfully?									
4.4 Filta-Max® filter wash station elution				Technician:					
4.4.1 Is an automatic or manual wash station used?									
4.4.2 Is the filter wash station set up correctly?	12.3.2.1	12.3.2.1	-	Requirement GLP					
4.4.3 Is residual suspension rinsed from all containers?	12.3.2.2.1d	12.3.2.2.1d	-	Critical					
4.4.4 Is PBST used to elute the filter?	7.4.2.4	7.6.2.4	-	Method Procedure					
4.4.5 Is an appropriate amount of PBST used for each wash? (approx. 600 mL)	12.3.2.2	12.3.2.2	-	Method Procedure					
4.4.6 During the first wash, is the plunger moved up and down 20 times?	12.3.2.2.1h	12.3.2.2.1h	-	Method Procedure					
4.4.7 Is the plunger moved up and down gently to avoid generating excess foam?	12.3.2.2.1h	12.3.2.2.1h	-	Method Procedure					
4.4.8 During the second wash, is the plunger moved up and down 10 times?	12.3.2.2.2b	12.3.2.2.2b	-	Method Procedure					
4.4.9 If the automatic washer is used, is the machine operating properly?	12.3.2.1	12.3.2.1	-	Requirement					
4.4.10 Is the wash station cleaned adequately between samples?	12.3.4.2	12.3.4.2	-	Requirement GLP					

Item to be evaluated	Reference*			Classification	Satisfactory				Comments/ Response Requested
	1623	1623.1	Cert		Yes	No	NA	UNK	
4.4.11 Are SOPs for Filta-Max® filter wash station elution available in the work area, and does laboratory practice reflect written procedures?				Critical GLP					
4.4.12 Other than issues noted for items 4.4.1 through 4.4.11 (if any) was Filta-Max® filter wash station elution demonstrated successfully?									
5 Concentration									
5.1 Filta-Max® filter sample concentration				Technician:					
5.1.1 Is concentrator set up correctly?	12.3.3.2.1b	12.3.3.2.1a	-	Requirement GLP					
5.1.2 Is the force of the vacuum maintained below 30 cm Hg?	NOTE pg 43	NOTE pg 34	-	Method Procedure					
5.1.3 Is concentration performed after each of the washes?	12.3.2.2.1j	12.3.2.2.1j	-	Method Procedure					
5.1.4 Is the sample concentrated so that some liquid remains above the filter (enough to cover the stir bar about half-way)?	12.3.3.2.1c	12.3.3.2.1b	-	Method Procedure					
5.1.5 Are the stir bar and concentration tube rinsed after each concentration and the liquid added to the concentrate?	12.3.3.2.1c	12.3.3.2.1b	-	Requirement					
5.1.6 Was the filter membrane washed twice with 5 mL of PBST?	12.3.3.2.3	12.3.3.2.3	-	Method Procedure					
5.1.7 Are SOPs for Filta-Max® filter sample concentration available in the work area, and does laboratory practice reflect written procedures?				Critical GLP					

Item to be evaluated	Reference*			Classification	Satisfactory				Comments/ Response Requested
	1623	1623.1	Cert		Yes	No	NA	UNK	
5.1.8 Other than issues noted for items 5.1.1 through 5.1.7 (if any) was Filta-Max® filter sample concentration demonstrated successfully?									
5.2 Envirochek® HV and Filta-Max® filter sample centrifugation				Technician:					
5.2.1 Is the sample centrifuged at 1500 x G (maximum 2000 x G) using a swinging bucket rotor?	13.2.1 and NOTE pg 46	13.2.1 and NOTE pg 37	-	Method Procedure GLP					
5.2.2 Are the centrifuge tubes properly balanced prior to centrifugation?	-	13.2.1	3.15.4	Critical					
5.2.3 Does lab have easily accessible method for determining relative centrifugal force of centrifuges?	-	-	3.15.1	Critical GLP					
5.2.4 Is the sample centrifuged for 15 minutes, with time beginning when centrifuge reaches desired speed?	13.2.1	13.2.1	-	Method Procedure					
5.2.5 Is the centrifuge slowly decelerated at the end without the brake?	13.2.1	13.2.1	-	Method Procedure					
5.2.6 Is the pellet volume determined?	13.2.1	13.2.1	5.2.3	Requirement					
5.2.7 Is there a set of standards for comparison of pellet size?	-	-	5.2.3	Recommendation GLP					
5.2.8 Are SOPs for Envirochek® and Filta-Max® filter sample centrifugation available in the work area, and does laboratory practice reflect written procedures?				Critical GLP					
5.2.9 Other than issues noted for items 5.2.1 through 5.2.8 (if any) was Envirochek® HV or Filta-Max® filter sample centrifugation demonstrated successfully?									

Item to be evaluated	Reference*			Classification	Satisfactory				Comments/ Response Requested
	1623	1623.1	Cert		Yes	No	NA	UNK	
6 Purification and Slide Preparation				Technician:					
6.1 Is an approved IMS kit/manufacture used?	7.5	7.7.1	-	Method Procedure GLP					
6.2 Is the supernatant from the centrifuged sample aspirated no lower than 5 mL of supernatant above every 0.5 mL pellet or portion of 0.5 mL pellet?	13.2.2	13.2.2 13.2.3	5.2.2 5.2.3	Requirement					
6.2.1 Are the samples aspirated using the pipette, with the documented internal diameter, as specified in the SOP?	-	NOTE pg 37	-	Critical					
6.2.2 Is the proper rate (mL/min) or pressure (psi) maintained throughout aspiration?	-	13.2.2 13.2.3	-	Method Procedure					
6.3 Is the pellet vortexed a sufficient time for resuspension?	13.2.3 13.2.4.1.3 13.2.4.2	13.2.2.1 13.2.3.1.2 13.2.3.2	-	Method Procedure					
6.4 Is the resuspended pellet volume quantitatively transferred to the flat-sided tube (2 rinses)?	13.3.2.1	13.3.2.1	-	Method Procedure					
6.5 Are the IMS beads thoroughly resuspended prior to addition to the flat-sided tube?	13.3.2.2 13.3.2.4	13.3.2.2 13.3.2.4	-	Method Procedure					
6.6 Is the flat-sided tube rotated at 18 rpm for 1 hour at room temperature?	13.3.2.6	13.3.2.6	-	Method Procedure					
6.7 Is the rotating mixer calibrated annually?	-	-	3.17.4	Critical GLP					
6.8 Is flat-sided tube correctly placed in magnet and rocked through 90 degrees about once per second?	13.3.2.7- 13.3.2.9	13.3.2.7- 13.3.2.9	-	Method Procedure					
6.9 Is all the liquid removed when decanting is performed with the magnet up?	13.3.2.11	13.3.2.11	-	Method Procedure					
6.10 Is the sample quantitatively transferred from the flat-sided tube to the microcentrifuge tube (2 rinses)?	13.3.2.13	13.3.2.14	-	Method Procedure					

Item to be evaluated	Reference*			Classification	Satisfactory				Comments/ Response Requested
	1623	1623.1	Cert		Yes	No	NA	UNK	
6.11 Are the beads rinsed with PBS while inside the microcentrifuge tube?	13.3.4	13.3.2.17	-	1623 Recommendation 1623.1 Requirement					
6.12 Is standard NaOH (5 µL, 1N) and standard HCl (50 µL, 0.1N) used?	NOTE pg 49 & 50	NOTE pg 42	3.17.5	Requirement GLP					
6.13 Is sample vortexed vigorously for 50 seconds immediately after the addition of acid and 30 seconds after the sample has set for 10 minutes at room temperature?	13.3.3.2- 13.3.3.4	13.3.3.2- 13.3.3.4	-	Method Procedure					
6.14 Is a second dissociation performed?	13.3.3.10 NOTE pg 49	13.3.3.10 NOTE pg 41	5.2.4	Requirement					
6.15 When the second dissociation is performed, does the laboratory: (A) use a second slide (B) add the additional volume to the original slide?	13.3.3.10	13.3.3.10 13.4.5	-	Circle one: A B					
6.16 Are the slides clearly labeled so they can be associated with the correct sample?	13.3.3.7	13.3.3.7	-	Requirement					
6.17 What type of slides is used?				GLP					
6.18 Is slide dried at: (A) room temperature, (B) 35° to 42°C, or (C) in the refrigerator?	13.3.3.12	13.3.3.12	-	Circle one: A B C					
6.19 If the slide is warmed, is incubator or slide warmer calibrated and labeled?	-	-	3.4	Critical GLP					
6.20 Are SOPs available in the work area for sample purification and slide preparation, and does laboratory practice reflect written procedures?				Critical GLP					
6.21 Other than issues noted for items 6.1 through 6.20 (if any) was purification and slide preparation demonstrated successfully?									
7 Sample Staining				Technician:					
7.1 What staining kit/manufacturer is used?	14.2	14.2	3.18.1	GLP					

Item to be evaluated	Reference*			Classification	Satisfactory				Comments/ Response Requested
	1623	1623.1	Cert		Yes	No	NA	UNK	
7.2 Is FITC stain applied according to manufacturer's directions?	14.2	14.2	5.3.2	Method Procedure					
7.3 Are positive and negative staining controls performed?	14.1	14.1	5.3.5	Requirement					
7.4 Are the slides incubated in a humid chamber in the dark at room temperature for approximately 30 minutes or per manufacturer's directions?	14.3	14.3	5.3.3	Method Procedure					
7.5 Are the labeling reagents rinsed away properly after incubation, without disturbing the sample?	14.5	14.5	-	Method Procedure					
7.6 Was the working DAPI stain prepared the day it was used?	7.7.2	7.9.2	3.19.2	Method Procedure					
7.7 Is stock DAPI stored at 1 to 10°C in the dark?	7.7.1	7.9.1	3.19.1	Method Procedure					
7.8 Is the DAPI stain applied properly and allowed to stand for a minimum of 1 minute?	14.6	14.6	-	Method Procedure					
7.9 Is the DAPI stain rinsed away properly without disturbing the sample?	14.7	14.7	-	Method Procedure					
7.10 Is the mounting media applied properly?	14.8	14.8	-	Method Procedure					
7.10.1 What type of mounting media is used?	7.8	7.10	-	GLP					
7.10.2 Are all the edges of the cover slip sealed well with clear fingernail polish, unless Elvanol® is used?	7.9 14.9	7.11 14.9	-	Method Procedure					
7.11 Are the finished slides stored in a humid chamber in the dark at 1 to 10°C (humid chamber not required for Elvanol®)?	14.10	14.10	5.3.6	Method Procedure					
7.12 Are SOPs for sample staining available in the work area, and does laboratory practice reflect written procedures?				Critical GLP					
7.13 Other than issues noted for items 7.1 through 7.12 (if any) was sample staining demonstrated successfully?									

Item to be evaluated	Reference*			Classification	Satisfactory				Comments/ Response Requested
	1623	1623.1	Cert		Yes	No	NA	UNK	
8 Microscope and Examination									
8.1 Is microscope equipped with appropriate excitation and band pass filters for examining FITC labeled specimens as demonstrated with lab, and auditor provided, positive staining control?	6.9.2	6.7.2	3.22.3	Requirement GLP					
8.2 Is microscope equipped with appropriate excitation and band pass filters for examining DAPI labeled specimens as demonstrated with lab, and auditor provided, positive staining control?	6.9.3	6.7.3	3.22.3	Requirement GLP					
8.3 Does the microscope have appropriate objectives and filters for DIC, which change easily to and from epifluorescence?	6.9.1	6.7.1	3.22.4	Requirement GLP					
8.4 Are all portions of the microscope, from the light sources to the oculars, properly adjusted?	10.3	10.0 Appendix B	3.22.6	Requirement					
8.5 Is the DIC image appropriate for each laboratory microscope?	-	Figure 4	Visual Guide	Requirement					
8.6 Is microscope cleaned after every session?	10.4	10.9.8	3.22.11	Requirement GLP					
8.7 Does the microscope have a 20X scanning objective?	6.9.1	6.7.1	3.22.8	Requirement GLP					
8.8 Does the microscope have a 100X oil immersion objective?	6.9.1	6.7.1	3.22.8	Requirement GLP					
8.9 Is the microscope equipped with an ocular micrometer?	6.9.1	6.7.1	3.22.9	Requirement GLP					
8.10 Is a stage micrometer available to laboratory?	6.9.1 10.3.5	6.7.1 App. B 3	3.22.9	Requirement					
8.11 Is a calibration table for 100X objective located close to the microscope(s)?	10.3.5.7	App. B 3.7	3.22.9	Requirement					
8.12 Has the mercury bulb been used less than the maximum hours recommended by the manufacturer?	10.3.2.11	App.B 1.11	3.22.12	Requirement					

Item to be evaluated	Reference*			Classification	Satisfactory				Comments/ Response Requested
	1623	1623.1	Cert		Yes	No	NA	UNK	
8.13 Does the laboratory have a preventative maintenance agreement in place to service the microscope annually?	-	-	3.22.6	Critical GLP					
8.14 Are SOPs for sample examination available in the work area, and does laboratory practice reflect written procedures?				Critical GLP					
8.15 Other than issues noted for items 8.1 through 8.13 (if any) was Microscope and Examination demonstrated successfully?									

9 Positive Staining Control and OPR Slides										
9.1	Does the laboratory's positive staining control slide contain (oo)cysts at the appropriate fluorescence intensity for FITC?	15.2.1.3	15.2.1.3	5.4.8 5.4.9.2 5.4.10.2	Requirement					
9.2	Does the laboratory's positive staining control slide contain (oo)cysts at the appropriate fluorescence intensity for DAPI?	15.2.1.3	15.2.1.3	5.4.8 5.4.9.3 5.4.10.3	Requirement					
9.3	Does the laboratory's positive staining control slide contain an appropriate level of background fluorescence?	-	-	5.4.3	Recommendation					
9.4	Is concentration of oocysts on the positive staining control slide appropriate?	14.1.1 15.2.1.3	14.1.1 15.2.1.3	7.1.8.1	Requirement					
9.5	Does the laboratory's positive staining control exhibit appropriate contrast and organism features by DIC?	-	Figure 4	Visual Guide	Requirement					
9.6	Does the laboratory's OPR slide contain (oo)cysts at the appropriate fluorescence intensity for FITC?	15.2.2.1 15.2.3.1	15.2.2.2 15.2.3.2	5.4.9.2 5.4.10.2	Requirement					
9.7	Does the laboratory's OPR slide contain (oo)cysts at the appropriate fluorescence intensity for DAPI?	15.2.2.2 15.2.3.2	15.2.2.3 15.2.3.3	5.4.9.3 5.4.10.3	Recommendation					
9.8	Does the laboratory's OPR slide contain an appropriate level of background fluorescence?	-	-	5.4.3	Requirement					
9.9	Does the laboratory's OPR slide exhibit appropriate contrast and organism features by DIC?	9.7.1.1	9.8.1.1 Figure 4	Visual Guide	Requirement					
9.10	Does the technical auditor's count of <i>Cryptosporidium</i> oocysts and <i>Giardia</i> cysts on the OPR slide sent by the laboratory agree within 10% of laboratory count?	10.6.3.1	9.10.3.1	7.1.9.4	Requirement					

Comments:

10 Onsite Sample Processing			
Method Step	Name	Position	Demonstrated Technique Successfully yes/no
Spiking – (filter type)			
Filtration - (filter type)			
Spiking flat-sided tube, and processing IMS control			
Aspiration and transfer from 250 mL bottle			

11 Onsite Blind Spike Results						
Sample	Crypto Spike Value	Crypto Count	Crypto Recovery (%)	Giardia Spike Value	Giardia Count	Giardia Recovery (%)

12 Evaluation of Onsite Sample Processing and Blind Spike Results – Comments and Recommendations		
Classification	Comments	Response Requested

13 Was analyst microscope operation acceptable? (yes/no)				
		Requirement <i>Method 1623: 10.3.4.1</i> <i>Method 1623.1: 10.7.1</i>	Requirement <i>Method 1623: 10.3.4.2-3</i> <i>Method 1623.1: 10.7.2-3</i>	Requirement <i>Method 1623: 10.3.6</i> <i>Method 1623.1: 10.8</i>
Name	Position	Adjust Interpupillary Distance	Focus both eyepieces	Establish Kohler Illumination

14 Slide Count and Analyst Verification Results (yes/no)					
	Requirement <i>Method 1623: 10.6.3.1</i> <i>Method 1623.1: 9.10.3.1</i>	Requirement <i>Method 1623: 10.6.3.1</i> <i>Method 1623.1: 9.10.3.1</i>	Requirement <i>Method 1623: 15.2</i> <i>Method 1623.1: 15.2</i>	Requirement <i>Method 1623: 15.2.2.3</i> <i>15.2.3.3</i> <i>Method 1623.1: 15.2.2.4</i> <i>15.2.3.4</i>	Requirement <i>Method 1623: 15.2.2.3</i> <i>15.2.3.3</i> <i>Method 1623.1: 15.2.2.4</i> <i>15.2.3.4</i>
Analyst	Crypto Count Within 10% of Target Count	<i>Giardia</i> Count Within 10% of Target Count	Examine and Record Characteristics	Measurement (100X)	Demonstrated Internal Structures

15 Evaluation of Analyst Microscopy and Examination Skills – Comments and Recommendations		
Classification	Comments	Response Requested

APPENDIX H

Example Positive Staining Control and OPR Slide Evaluation

Laboratory Name:			Date:			
Technical Auditor:			Organism/Slide ID: <i>Cryptosporidium</i>			
		1	2	3	4	Score
Positive Stain Control	Number of Organisms [Sections 14.1.1 & 15.2.1.3 (Sections 14.1.1 & 15.2.1.3)]	organisms in very low numbers (<~25 oo/cysts)	organisms in low numbers (~25 to ~100 oo/cysts)	organisms in relatively high numbers(~100 to <~200 oo/cysts)	organisms in high numbers (>~200 oo/cysts)	
	FITC Fluorescence* [Section 15.2.1.3 (Section 15.2.1.3)]	consistently weak (1+); inadequate; not crisp	inconsistent; more weak (1+) than strong (3+); less than adequate; more not crisp than crisp	inconsistent; more strong (3+) than weak (1+); adequate; more crisp than not crisp	consistently strong (3+); superior; crisp	
	DAPI Fluorescence** [Section 15.2.1.3 (Section 15.2.1.3)]	DAPI negative in majority of oocysts; nuclei stained consistently weak (1+); inadequate	distinct nuclei in less than half of the oocysts; nuclei stain inconsistent; more weak (1+) than strong (3+); less than adequate	distinct nuclei stained in majority of oocysts; nuclei stain inconsistent; more strong (3+) than weak (1+); adequate	distinct nuclei stained in majority of oocysts; nuclei stain consistently strong (3+); superior	
	Background Fluorescence	excessive, interfering	distracting	exists but does not interfere	minimal to nonexistent	
	Background and Organism Characteristics on DIC	interfering; atypical	distracting; atypical	exists but does not interfere; typical	minimal to nonexistent; typical	

OPR	Number of Organisms [Section 10.6.3.1 (Section 9.10.3.10)]	difference in number counted >20%	16-20% difference	10-15% difference	difference in number counted <10%	
	FITC Fluorescence* [Section 15.2.2.1 (Section 15.2.2.2)]	consistently weak (1+); inadequate; not crisp	inconsistent; more weak (1+) than strong (3+); less than adequate; more not crisp than crisp	inconsistent; more strong (3+) than weak (1+); adequate; more crisp than not crisp	consistently strong (3+); superior; crisp	
	DAPI Fluorescence** [Section 15.2.2.2 (Section 15.2.2.3)]	DAPI negative in majority of oocysts; nuclei stained consistently weak (1+), inadequate	distinct nuclei in less than half of the oocysts; nuclei stain inconsistent; more weak (1+) than strong (3+); less than adequate	distinct nuclei stained in majority of oocysts; nuclei stain inconsistent; more strong (3+) than weak (1+); adequate	distinct nuclei stained in majority of oocysts; nuclei stain consistently strong (3+), superior	
	Background Fluorescence	excessive, interfering	distracting	exists but does not interfere	minimal to nonexistent	
	Background and Organism Characteristics on DIC [[Figure 4]]	interfering; <50% of oo/cysts undamaged and morphologically intact	distracting; <50% oo/cysts undamaged and morphologically intact	exists but does not interfere; >50% oo/cysts undamaged and morphologically intact	minimal to nonexistent; >50% oo/cysts undamaged and morphologically intact	

*FITC Fluorescence: brilliant apple-green ovoid or spherical objects with brightly highlighted edges (referred to as crisp above); compared for all organisms over complete well slide at 200x
**DAPI Fluorescence: light blue internal staining (no distinct nuclei) with green rim, intense blue internal staining, or distinct, sky-blue nuclei; compared for all organisms over complete well slide at minimum of 400x
Fluorescence intensity scale: 1+ = weak, 2+=medium, 3+=strong
For visual comparisons, see FITC and DAPI staining examples in Method 1623.1 Microscopy Visual Guide (p. 61 in Method 1623.1) or the LT2 On-line Microscopy Training Module (<http://water.epa.gov/lawsregs/rulesregs/sdwa/lt2/compliance.cfm#training>). For descriptions, see Method 1623/1623.1 Sections 15.2.2 and 15.2.3.
Section references in [] refer to EPA Method 1623, and the corresponding section references in parentheses and italics refer to EPA Method 1623.1.

Satisfactory scores are ≥ 3. Any score ≤ 2 (shown in red font) indicates problems with slide preparation and should be investigated. If slide evaluation is conducted as part of the Assessment of *Cryptosporidium* Laboratory Quality Assurance under the Safe Drinking Water Act, scores, comments and any requirements, recommendations, or commendations should be included in Section 9 of Checklist C - Method 1623/1623.1 Technical Review - Sample Processing and Microscopy.

Comments:
PSC:
OPR:

Requirements, Recommendations, Commendations:

APPENDIX I

Example Report from Online Analyst Evaluation

APPENDIX J

Method 1623 Ongoing Precision and Recovery (OPR) and Initial Precision and Recovery (IPR)
Quality Control Criteria Calculated from Five Rounds of Proficiency Testing (PT) Data

Method 1623 Ongoing Precision and Recovery (OPR) and Initial Precision and Recovery (IPR) Quality Control Criteria Calculated from Five Rounds of Proficiency Testing (PT) Data

Updated July 9, 2013

Introduction

The December 2005 Method 1623 Ongoing Precision and Recovery (OPR) and Initial Precision and Recovery (IPR) quality control and acceptance criteria were developed based on 293 *Cryptosporidium* OPR samples and 186 *Giardia* OPR samples analyzed by six laboratories during the Information Collection Rule Supplemental Surveys from March 1999 to February 2000. In 2009, the minimum *Cryptosporidium* percent recovery for OPR samples was updated based on analysis of data from the February and May 2007 proficiency testing (PT) rounds as published in the Federal Register ([Federal Register Notice, Vol. 74, No. 36 February 25, 2009](#)).

To more accurately reflect current laboratory capability and provide better values for assessment of laboratory performance, data from five PT rounds (February and May 2007, July 2009, February 2010, and March 2011) were used to develop quality control criteria. For each round, laboratories analyzed three blind reagent water samples spiked with flow-cytometer counted organisms prepared by the Wisconsin State Laboratory of Hygiene. These updated criteria provide a better assessment of laboratory capability using Method 1623 than the 2009 values for the following reasons: 1) the data set is more recent; and 2) the sample size is more than twice as large as the 2009 sample size. A summary of the data set and proposed acceptance criteria are shown in Table 1.

Calculation of OPR Minimum Recovery Criteria

The minimum *Cryptosporidium* percent recovery criteria for OPR samples were calculated based on data from the February and May 2007, July 2009, February 2010, and March 2011 PT rounds, which were all of the PT rounds with protozoan suspensions in reagent water without any additional matrix material. The remaining PT rounds had matrix additions and would not simulate an OPR or IPR. The acceptance criteria were calculated using data from labs approved and participating in the Lab QA Program as of June 24, 2011. The calculated lower limit of recovery was 33% for *Cryptosporidium* and 29% for *Giardia*. When applying the criteria to the data from the five PT rounds, 51 samples or 6.8% of the 753 samples did not meet the criterion for *Cryptosporidium*, and 31 samples, or 5.8% of the 530 samples, did not meet the criterion for *Giardia*.

Details of the Calculation of OPR Minimum Recovery Criteria

To calculate OPR recovery criteria, estimates of variance attributable to four different sources were calculated: variability between laboratories, variability between PT rounds, variability between laboratory-and-round (i.e., attributable to an interaction between round and laboratory), and variability within round and laboratory (i.e., analytical variability). There were a few laboratories that performed PT analyses using multiple filters. For the purpose of these calculations, the two sets of analyses were two separate entities; in other words, the two lab/filter combinations were treated as two different laboratories. The different variance components were calculated using PROC MIXED from SAS version 8 using the maximum likelihood method of

estimation on recovery data. Details on the maximum likelihood estimation can be found in SAS/STAT User's Guide.¹

Estimates of between laboratory variance, between round variance, between laboratory-and-round variance, and within laboratory-and-round variance were labeled s^2_L , s^2_R , s^2_{LR} and s^2_w , respectively.

The combined standard deviation (s_c) is:

$$s_c = \sqrt{\left(1 + \frac{1}{\bar{L}}\right) s^2_L + \left(1 + \frac{1}{\bar{R}}\right) s^2_R + \left(1 + \frac{1}{\bar{L} * \bar{R}}\right) s^2_{LR} + \left(1 + \frac{1}{\bar{L} * \bar{R} * n}\right) s^2_w}$$

Where:

$$\bar{L} = \frac{C}{R} \quad = \text{average number of laboratory/filter combinations per round}$$

$$\bar{R} = \frac{C}{L} \quad = \text{average number of rounds per laboratory/filter combination}$$

n = average number of replicates per laboratory/filter and round (3 for *Cryptosporidium*, 2.2 for *Giardia*)

C = total number of laboratories/filters/rounds

R = number of rounds (5)

L = number of laboratory/filter combinations

Lower recovery limits for OPR samples were then calculated as:

$$\bar{X}_{Mean} - t_{(0.95, df)} * s_c$$

Where:

\bar{X}_{mean} = the mean recovery of all samples

$t_{(0.95, df)}$ = 95th percentile of the student's t distribution with df degrees of freedom
 df is calculated using Satterthwaite's estimate as given below:

$$df = \frac{s_c^4}{\frac{\left(\left(1 + \frac{1}{\bar{L}}\right) s^2_L\right)^2}{\bar{L} - 1} + \frac{\left(\left(1 + \frac{1}{\bar{R}}\right) s^2_R\right)^2}{\bar{R} - 1} + \frac{\left(\left(1 + \frac{1}{\bar{L} * \bar{R}}\right) s^2_{LR}\right)^2}{(\bar{L} - 1)(\bar{R} - 1)} + \frac{\left(\left(1 + \frac{1}{\bar{L} * \bar{R} * n}\right) s^2_w\right)^2}{\bar{L} * \bar{R} * (n - 1)}}$$

Calculation of IPR Minimum Mean Recovery and Maximum RSD Criteria

The QC acceptance criteria for IPR samples were calculated based on data from the February and May 2007, July 2009, February 2010, and March 2011 PT rounds. The calculated lower limit of recovery was 36% for *Cryptosporidium* and 32% for *Giardia*. When applying the criteria to the data from the five PT rounds, 16 (6.4%) of the 251 round/filter/laboratory combinations did not meet the criterion for minimum mean recovery for *Cryptosporidium*, and 17 (7.1%) of the 240 combinations did not meet the minimum mean recovery criterion for *Giardia*. The maximum RSD of 34% for *Cryptosporidium* and 37% for *Giardia* was also calculated using data from the five PT rounds. Applying the criteria to the data from all laboratories, 14 laboratory/filter/round combinations (5.6%) did not meet the *Cryptosporidium* criterion and 14 combinations (5.8%) did not meet the *Giardia* criterion. Six of the 16 *Cryptosporidium* lab/round/filter combinations and 5 of the 17 *Giardia* combinations that failed the lower recovery criterion also failed the precision criterion. It is worth noting that because the

¹ SAS Institute Inc. 1994. SAS/STAT User's Guide, Volume 2, GLM-VARCOMP. Version 6, 4th Edition, June 1994.

criteria are calculated for 4 samples, while the data included only 3 (*Cryptosporidium*) or 2 (*Giardia*) results per lab, the failure rates would be expected to be greater than the target 5%. However, due to the large number of laboratory/filter/round combinations, the failure rate was ≈5%.

Details of the Calculation of IPR Criteria

Similar to the calculations for the OPR recovery criteria, IPR recovery criteria were calculated with estimates of variance attributable to four different sources: variability between laboratories, variability between PT rounds, variability between laboratory-and-round (i.e., attributable to an interaction between round and laboratory), and variability within round and laboratory (i.e., analytical variability). There were a few laboratories that performed PT analyses using multiple filters. For the purpose of these calculations, the two sets of analyses were two separate entities; in other words, the two lab/filter combinations were treated as two different laboratories. The different variance components were calculated using PROC MIXED from SAS version 8 using the maximum likelihood method of estimation on recovery data. Details on the maximum likelihood estimation can be found in SAS/STAT User's Guide.²

Estimates of between laboratory variance, between round variance, between laboratory-and-round variance, and within laboratory-and-round variance were labeled s^2_L , s^2_R , s^2_{LR} and s^2_w , respectively.

The combined standard deviation (s_c) is:

$$s_c = \sqrt{\left(1 + \frac{1}{\bar{L}}\right)s^2_L + \left(1 + \frac{1}{\bar{R}}\right)s^2_R + \left(1 + \frac{1}{\bar{L} * \bar{R}}\right)s^2_{LR} + \left(\frac{1}{4} + \frac{1}{\bar{L} * \bar{R} * n}\right)s^2_w}$$

Where:

$$\bar{L} = \frac{C}{R} \quad = \text{average number of laboratory/filter combinations per round}$$

$$\bar{R} = \frac{C}{L} \quad = \text{average number of rounds per laboratory/filter combination}$$

n = average number of replicates per laboratory/filter and round (3 for *Cryptosporidium*, 2.2 for *Giardia*)

C = total number of laboratories/filters/rounds

R = number of rounds (5)

L = number of laboratory/filter combinations

Lower recovery limits for the mean of IPR sample results were then calculated as:

$$\bar{X}_{Mean} - t_{(0.95, df)} * s_c$$

Where:

\bar{X}_{mean} = the mean recovery of all samples

$t_{(0.95, df)}$ = 95th percentile of the student's t distribution with df degrees of freedom

df is calculated using Satterthwaite's estimate as given below:

$$df = \frac{s_c^4}{\frac{\left(\left(1 + \frac{1}{\bar{L}}\right)s^2_L\right)^2}{\bar{L} - 1} + \frac{\left(\left(1 + \frac{1}{\bar{R}}\right)s^2_R\right)^2}{\bar{R} - 1} + \frac{\left(\left(1 + \frac{1}{\bar{L} * \bar{R}}\right)s^2_{LR}\right)^2}{(\bar{L} - 1)(\bar{R} - 1)} + \frac{\left(\left(\frac{1}{4} + \frac{1}{\bar{L} * \bar{R} * n}\right)s^2_w\right)^2}{\bar{L} * \bar{R} * (n - 1)}}$$

² SAS Institute Inc. 1994. SAS/STAT User's Guide, Volume 2, GLM-VARCOMP. Version 6, 4th Edition, June 1994.

The precision criterion for IPR samples was calculated as a maximum relative standard deviation (RSD). The maximum RSD was determined by first pooling the individual laboratory/round RSDs using the formula below:

$$RSD_{pool} = \sqrt{\frac{1}{C} \sum_{i=1}^C RSD_i^2}$$

Where:

RSD_i = the RSD calculated for laboratory/filter/round i , and

C = the number of laboratory/filter/round combinations.

The maximum RSD was then calculated as:

$$RSD_{Max} = \sqrt{F_{(0.95;3,n_T-C)}} * RSD_{Pool}$$

Where:

n_T = the total number of results from all laboratories over all rounds, and

C = the number of laboratory/filter/round combinations.

Table 1. Summary of Acceptance Criteria

	Method 1623 2005 version	Federal Register 74:36 (February 25, 2009) p. 8529	Updated Value
OPR Criteria – Lower Limit	<i>Cryptosporidium</i> - 11% <i>Giardia</i> - 14%	<i>Cryptosporidium</i> - 22%	<i>Cryptosporidium</i> - 33% <i>Giardia</i> - 29%
IPR Criteria – Lower Limit	<i>Cryptosporidium</i> - 24% <i>Giardia</i> - 24%	NA	<i>Cryptosporidium</i> - 36% <i>Giardia</i> - 32%
IPR Criteria – Precision (maximum RSD)	<i>Cryptosporidium</i> - 55% <i>Giardia</i> - 49%	NA	<i>Cryptosporidium</i> - 34% <i>Giardia</i> - 37%
Data Set	ICRSS data generated in 1999 and 2000	PT data generated during the February and May 2007 rounds	PT data generated, as of June 24, 2011, during five PT rounds
Number of Laboratories/Filters	6	58	<i>Cryptosporidium</i> - 56 <i>Giardia</i> - 55
Number of Samples	293	333	<i>Cryptosporidium</i> - 753 <i>Giardia</i> - 530
Method Version Used	1999 version of Method 1623 – Envirochek only	2005 version of Method 1623 – all filters	2005 version of Method 1623 – all filters
Blind vs. Unblind	Unblind	Blind	Blind

APPENDIX K

Burden Tables

Table 1. Participating Laboratories Seeking Certification

Activity	Legal \$65.00/hour	Management \$98.00/hour	Technical \$82.00/hour	Clerical \$72.00/hour	Respondent Hours	Labor Costs	Capital/Start up	O&M Costs	Number of respondents/year	Total hours/year	Total cost/year	Labor Cost/ Activity	O&M Cost/Activity	Labor Cost/Hr
Complete and submit laboratory evaluation application package (Rpt, TPD - Start up and O&M costs)	0	3	10	1	13				22.5	293				
	\$ -	\$ 257.25	\$ 799.50	\$ 48.00		\$ 1,104.75	\$ -	\$3,920.00			\$113,056.88	\$24,856.88	\$ 88,200.00	\$ 84.71
Perform off-site evaluation activities (OPR/PSC evaluation, online analyst verification) (Rpt)	0	4	18	0	22				22.5	486				
	\$ -	\$ 367.50	\$ 1,435.00	\$ 24.00		\$ 1,826.50	\$ -	\$700.00			\$ 56,846.25	\$41,096.25	\$ 15,750.00	\$ 84.63
Host on-site evaluation (includes counting slides) (Rpt; TPD - O&M costs)	0	14	24	2	40				22.5	889				
	\$ -	\$ 1,323.00	\$ 1,968.00	\$ 144.00		\$ 3,435.00	\$ -	\$3,254.00			\$150,502.50	\$77,287.50	\$ 73,215.00	\$ 86.96
Perform and report 2 sets of proficiency tests (PT) (Rpt)	0	4	29	2	34				45	1541				
	\$ -	\$ 343.00	\$ 2,357.50	\$ 144.00		\$ 2,844.50	\$ -	\$2,225.00			\$228,127.50	\$128,002.50	\$100,125.00	\$ 83.05
Perform follow-up action for poor PT results (Rpt)	0	3	19	1	23				7	158				
	\$ -	\$ 294.00	\$ 1,517.00	\$ 72.00		\$ 1,883.00	\$ -	\$1,266.67			\$ 22,047.67	\$13,181.00	\$ 8,866.67	\$ 83.69
Perform and report 2 sets of PTs for each additional method version (Rpt)	0	4	29	2	34				4	137				
	\$ -	\$ 343.00	\$ 2,357.50	\$ 144.00		\$ 2,844.50	\$ -	\$2,225.00			\$ 20,278.00	\$11,378.00	\$ 8,900.00	\$ 83.05
Total									3504	3504	\$590,858.79	\$295,802.13	\$295,056.67	

O&M Costs to Complete and submit laboratory evaluation application package: application fee (average value from 2 lab and 2 states)
 O&M Costs to Perform off-site evaluation activities: slide production, cost of shipping slides to States/EPA for evaluation
 O&M Costs to Host on-site evaluation: cost of audit package including slides, travel costs for auditor
 O&M Costs to Perform and report 2 sets of proficiency tests (PT): cost of PT samples and supplies
 O&M Costs to Perform follow-up action for poor PT results: cost of additional blind samples and supplies
 O&M Costs to Perform and report 2 sets of PTs for each additional method version: cost of PT samples and supplies
 Number of respondents/year to Complete and submit audit package: Total of approximately 45 laboratories seeking approval divided by two years
 Number of respondents/year to Perform off-site evaluation activities: Total of approximately 45 laboratories seeking approval divided by two years
 Number of respondents/year to Host on-site evaluation: Total of approximately 45 laboratories seeking approval divided by two years
 Number of respondents/year to Perform and report 2 sets of proficiency tests (PT): Approximately 45 labs participating in the Lab QA Program
 Number of respondents/year to Perform follow-up action for poor PT results : Estimated 7 laboratories will have to perform follow-up due to poor PT results
 Number of respondents/year to Perform and report 2 sets of PTs for each additional method version: Estimated 4 laboratories will perform more than one method version

Rpt - Reporting
 RK - Record keeping
 TPD - Third-party disclosure

Table 2. State Annual Burden

Activity	Certification Officer/ Technical \$107/hr	Management \$120/hr	States hrs/yr/resp	Labor cost/yr/resp	Capital Startup Cost	O&M Costs	Number of Labs	Total hrs/yr	Total Costs per Year	Labor Cost/Activity	O&M Cost/Activity
Review laboratory evaluation packages (RK)	16.5	2	18.5				22.5	416			
	\$ 1,765.50	\$ 240.00		\$ 2,005.50		\$ -			\$ 45,123.75	\$45,123.75	\$ -
Conduct and review off-site evaluation activities (Rpt)	12	2	14				22.5	315			
	\$ 1,284.00	\$ 240.00		\$ 1,524.00		\$ -			\$ 34,290.00	\$34,290.00	\$ -
Conduct and review on-site evaluations (Rpt)	18	2	20				22.5	450			
	\$ 1,926.00	\$ 240.00		\$ 2,166.00		\$ -			\$ 48,735.00	\$48,735.00	\$ -
Notify laboratories of certification status and maintain certified laboratory list on State website (Rpt)	25.5	2	27.5				22.5	619			
	\$ 2,728.50	\$ 240.00		\$ 2,968.50		\$ -			\$ 66,791.25	\$66,791.25	\$ -
Review and track 2 sets of PT results (RK)	3		3				45	135			
	\$ 321.00	\$ -		\$ 321.00		\$ -			\$ 14,445.00	\$14,445.00	\$ -
Coordinate follow-up activities for poor PT results	3		3				7	21			
	\$ 321.00	\$ -		\$ 321.00		\$ -			\$ 2,247.00	\$2,247.00	\$ -
Review and track 2 sets of PT results for additional method versions (RK)	3		3				4	12			
	\$ 321.00	\$ -		\$ 321.00		\$ -			\$ 1,284.00	\$1,284.00	\$ -
Total							1968		\$ 212,916.00	\$212,916.00	-

Information for Legal, Clerical, and Expert/Contractor personnel categories was not included because consultations revealed those personnel were not applicable.

O&M Costs: none

Number of respondents/year to Review laboratory evaluation packages: Total of approximately 45 laboratories seeking certification divided by two years

Number of respondents/year to Conduct and review off-site evaluation activities: Total of approximately 45 laboratories seeking certification divided by two years

Number of respondents/year to Conduct and review on-site evaluations: Total of approximately 45 laboratories seeking certification divided by two years

Number of respondents/year to Notify laboratories of certification status: Total of approximately 45 laboratories seeking certification divided by two years

Number of respondents/year to Review and track PT results (PT): Approximately 45 labs participating in the Lab QA Program

Number of respondents/year to Coordinate follow-up activities for poor PT results: Estimated 7 laboratories will have to perform follow-up due to poor PT results

Number of respondents/year to Review and track 2 sets of PT results for additional method versions: Estimated 4 laboratories will be performing more than one method version

Rpt - Reporting

RK - Record keeping

TPD - Third-party disclosure

Table 3. Agency Annual Burden

Activity	Program Management GS 14 \$87.25/hr	Management \$62.60/hr	Technical \$43.57/hr	Agency hrs/yr/resp	Labor cost/yr/resp	Capital Startup Cost	O&M Costs	Number of Laboratories	Total hrs/yr	Total Costs per Year	Labor Cost/Activity	O&M Cost/Activity
Assist State Certification Officers (CO) - laboratory evaluation application packages	5	0.5	15	20.5				22.5	461			
	\$ 436.25	\$ 31.30	\$ 653.55		\$ 1,121.10		\$ -			\$ 25,224.75	\$25,224.75	\$ -
Assist State Certification Officers (CO) - off-site evaluation activities (OPR/PSC evaluation, online analyst verification)	5	0.5	12	17.5				22.5	394			
	\$ 436.25	\$ 31.30	\$ 522.84		\$ 990.39		\$1,775.00			\$ 24,058.78	\$22,283.78	\$ 1,775.00
Assist State Certification Officers (CO) - on-site evaluation activities	10	0.5	8	18.5				22.5	416			
	\$ 872.50	\$ 31.30	\$ 348.56		\$ 1,252.36		\$ -			\$ 28,178.10	\$28,178.10	\$ -
Assist State Certification Officers (CO) - interpretation of PT results (2 sets/year)	7	0.25	4	11.25				45.0	506			
	\$ 610.75	\$ 15.65	\$ 174.28		\$ 800.68		\$ -			\$ 36,030.60	\$36,030.60	\$ -
Assist State Certification Officers (CO) - interpretation of PT results for additional method versions (2 sets/year)	7	0.25	4	11.25				4.0	45			
	\$ 610.75	\$ 15.65	\$ 174.28		\$ 800.68		\$ -			\$ 3,202.72	\$3,202.72	\$ -
Maintain links to State's websites for lists of certified laboratories	3	0.5	3	7				20.0	130			
	\$ 261.75	\$ 31.30	\$ 130.71		\$ 423.76		\$ -			\$ 8,475.20	\$8,475.20	\$ -
Total				1953						\$ 125,170.15	\$123,395.15	\$1,775.00

Information for Legal, Clerical, and Expert/Contractor personnel categories was not included because consultations revealed those personnel were not applicable. Costs and hours reflect 45 labs seeking certification once every 2 years (45/2=22.5), 45 labs completing 2 PT rounds per year, and 20 States with certification programs
O&M Costs: cost for software license (Adobe Connect) and annual microscope service

Table 4a. Total Respondent Burden

	Number of respondents	Number of Activities	Total hours/year	Total Labor cost/year	Total Annual Capital costs	Total Annual O&M Costs	Total Annualized Cost
Laboratories	45	6	3504	\$ 295,802.13	\$ -	\$ 295,056.67	\$ 590,858.79
States	20	7	1968	\$ 212,916.00	\$ -	\$ -	\$ 212,916.00
Total Burden	65	13	5472	\$ 508,718.13	\$ -	\$ 295,056.67	\$ 803,774.79

Table 4b. Total Agency Burden

	Number of respondents	Number of Activities	Total hours/year	Total Labor cost/year	Total Annual Capital costs	Total Annual O&M Costs	Total Annualized Cost
Total Burden	1	6	1953	\$ 123,395.15	\$ -	\$ 1,775.00	\$ 125,170.15