ATTACHMENT A

Analytical Laboratory Standard Operating Procedures and QAPP

Effective September 30, 2014
(This document supersedes that which was effective July 2013)

LABORATORY QUALITY MANAGEMENT PLAN/QUALITY ASSURANCE MANUAL

Of

BABCOCK LABORATORIES, INC.
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***Note: Noted appendices and referenced SOPs are available upon request.

1.0 INTRODUCTION

This document outlines the Quality Management Plan and Quality Assurance procedures implemented by Babcock Laboratories, Inc., a privately owned laboratory involved primarily in the testing of drinking water, wastewater, soils, and other matrices including food products and bottled beverages. (http://www.babcocklabs.com). This document may also be referred to as the Laboratory Management System per ISO 17025 requirements. This document describes the framework by which the laboratory establishes and maintains a documented quality system appropriate to the type, range, and volume of environmental and food/beverage testing activities it undertakes. This document outlines the laboratory's policies and procedures established in order to meet the requirements set by The Nelac Institute (TNI) for the National Environmental Laboratory Accreditation Program (NELAP), the Environmental Laboratory Accreditation Program (ELAP) for the state of California, the ISO/IEC 17025:2005 Standard "General requirements for the competence of testing and calibration laboratories", the requirements of the "AOAC" food program and the EPA Requirements for Quality Management Plans, EPA QA/R-2. See Appendix A for copies of the laboratory's certifications. Other documents and procedures will be referenced and should be consulted for specific details. For advertising purposes the use of the term "A2LA" and the "A2LA Accredited" symbol requires prior approval of QA and will be done in strict accordance with A2LA's advertising policy document (Appendix B). The Quality and Management Plan is maintained to provide accurate and dependable data for the laboratory's clients. It is the responsibility of each employee to be familiar with and implement the quality control practices of the laboratory. Management assures understanding and implementation of quality practices through all areas of the company and staff through training, Standard Operating Procedures, method and system audits, staff meetings, etc. Additionally, management is responsible for maintaining Quality Assurance (QA) in all aspects of the operation.

2.0 COMPANY VISION AND MISSION STATEMENT

2.1 Company Vision: *Creating a positive environment where laboratory professionals proudly deliver unparalleled value.*

2.2 Mission: *The goal of Babcock Laboratories, Inc. is to produce the highest quality, most reliable environmental services and analytical data.*

3.0 OBJECTIVES

3.1 To achieve this goal the corporate officers, board of directors and managers of the company are committed to these objectives:

3.1.1 To provide professional service to clients and the community, drawing on the many years of experience in the analytical testing industry.

3.1.2 To produce scientifically valid and legally defensible data.

3.1.3 To promote ethical standards and professional integrity within our organization and the environmental community.

3.1.4 To attract and retain the best, well educated, and properly trained staff possible.

3.1.5 To promote growth of the company in a fiscally responsible and profitable manner.

4.0 FLOW OF RESPONSIBILITY:

4.1 The flow of responsibility within the laboratory follows the organizational chart. See Appendix C for Resumes of Key Personnel and Appendix D for Organization Charts. Job descriptions for several key positions are attached to this document in Appendix E. Job descriptions discuss roles, responsibilities and authority of each position. Job descriptions for all positions are maintained in the personnel files.

4.1.1 The CEO is responsible for the business functions of the company.

4.1.2 The CEO oversees the office and the generation of all final reports.

4.1.3 The CFO oversees Finance, Accounting, Human Resources and Safety.

4.1.4 The Laboratory Director of Operations is in control of the operations of the laboratory. The Laboratory Technical Director is in control of instrumentation, methodology and technical areas of the laboratory.

4.1.5 The QA Manager has access to the CEO and Laboratory Directors and is responsible for ensuring lab wide compliance with California ELAP requirements, the TNI standards, the ISO 17025 standard and the EPA QA/R-2. The QA Manager reports to the CEO.

4.1.6 The Assistant QA Manager/QA Auditor and QA Officer help the QA Manager with all areas of the company's Quality Assurance Program and requirements.

4.1.7 The Project Managers are responsible for ensuring that client needs are met with respect to QC requirements and final reporting. Project Managers are also approved signatories for client reports.

4.1.8 The Inorganic Department Manager oversees the Inorganic Department. The Metals/IC Manager oversees the Trace Metals and Ion Chromatography Departments. The Organic Manager oversees the Volatile and Semi-Volatile Departments. The Microbiology Manager oversees the Microbiology Department. The Field Manager oversees the Field Department.

4.1.9 Managers are accountable for the smooth operation of their sections, meeting holding time deadlines, and troubleshooting problems. They ensure the competence of all who operate specific equipment, perform analytical tests, evaluate results, and sign test reports.

4.1.10 The chemists and technicians are held responsible for following prescribed protocols in the performance of their assigned analyses, and keeping their managers apprised of any difficulties

that could affect the accuracy of results or the smooth operation of the laboratory.

4.1.11 Where contracted and additional technical and key support personnel are used, the laboratory ensures that such personnel are supervised and competent and that they work in accordance with the laboratory's quality system.

4.2 The CEO is named as Deputy Laboratory Technical or Operations Director and fulfills that responsibility when one or both of the Laboratory Directors are absent. See Appendix E for a list of specific duties. If this absence exceeds 65 consecutive calendar days, CADHS-ELAP will be notified in writing. The Laboratory Director(s) are named as Deputy QA Officers and fulfill that responsibility when the QA Manager and Assistant QA Manager are absent.

4.3 The QA Department includes the QA Manager and designee(s) (i.e. Assistant QA Manager/QA Auditor and QA Officer) who perform tasks under the direction of the QA Manager. The members of the QA Department may also be referred to as QA Officers. The QA Department is part of the Administration Department and is independent of laboratory operations including groups responsible for the generation, compilation and evaluation of environmental data. The QA Office is located in the QA/Accounting/Finance office. QA files are maintained in the QA Office.

5.0 Contract Review

5.1 The laboratory has established procedures for the review of requests, tenders and contracts. Before the initiation of any analytical testing program, a review of the request is required.

Contract Review is handled by the Regional Business Development Director or Associate, the CEO and CFO, in collaboration with the Director of Client Services, the Client Services Manager and the Laboratory Director(s). Records of reviews are maintained for ongoing work, along with any pertinent information regarding client's requirements. An oral agreement or a written contract may be entered into to provide the client with testing services. Any differences between the request or tender and the contract shall be resolved before work is commenced. The client is to be contacted if laboratory accreditation is suspended, revoked, or voluntarily withdrawn. For more information on contract review, refer to the Review of Contracts and Tenders SOP (A13).

5.2 Laboratory Capacity/Planning

5.2.1 It is the responsibility of the Laboratory Director(s) to ensure adequate capacity for all new projects prior to commencing work. The system used to determine laboratory capacity is based on the time taken to analyze a batch of samples for a given analyte, the number of analysts and labor hours available, and the equipment at hand. Each test is monitored for frequency of request on a weekly basis. When new work is contemplated, workloads are reviewed to determine what number of samples can be added while remaining within the laboratory capacity to perform the work. In addition, management, QA and project management often discuss upcoming projects to ensure that client specific project requirements, methods, QC requirements, reporting formats, electronic reporting requests, etc. can be met. All project information and client requirements are noted in the LIMs system as well as Goldmine (Client database software). For more information on laboratory planning, refer to SOP A13.

6.0 CONFIDENTIALITY

6.1 **Laboratory reports** and accompanying documents contain confidential information intended for use by the individual or entity requesting and purchasing the analytical services. Except when required by law, no information relating to a report is released to another person or party without permission from the paying client.

6.2 Laboratory documents containing client's information are to be handled and disposed of in a discrete manner.

6.3 Permission to release information may be given by **telephone or in writing**. Documentation of consent, including the name of the person and the date/time of consent, is recorded in the client file.

6.4 **Faxes** clearly identify the intended recipient. The fax coversheet utilized by Babcock Laboratories contains a statement at the bottom of the sheet stating that the contents of the fax are confidential and intended only for the recipient. If the laboratory is familiar with a particular client, and there is agreement between the lab and the client, the cover letter may be omitted for brevity purposes.

6.5 Results transmitted via **E-mail or EDD** are also accompanied by a confidentiality statement.

6.6 Client results may be accessed via the **Internet**. These results are password protected and hosted by a server separate from the laboratory's main server.

6.7 Employees are informed of these policies during new employee orientation.

7.0 EXCEPTIONAL CIRCUMSTANCES

7.1 In the event that it is necessary to deviate from a documented policy, procedure or specification, several steps must be taken for approval of the exception. The QA Manager and Laboratory Directors meet to discuss and research the proposed exception. When circumstances are such that the QA Manager and Lab Directors agree that permission to deviate from policy, procedure or specification is warranted, the following steps must be taken.

7.1.1 Where applicable, the client is contacted for approval of the proposed change in procedure and verbal or written approval is requested and receipt verified.

7.1.2 The agreed upon change in policy, procedure, or specification is documented and kept in the project file.

7.1.3 Copies of the change are attached to the analytical records, where applicable.

7.1.4 Copies of the change are attached to the review reports for any analyses directly related to the change.

7.1.5 Copies of the change are filed in the client report file.

8.0 COMPLAINTS

8.1 Client concerns regarding any aspect of laboratory services are directed to the Director of Client Services, the Client Services Manager, the Regional Business Development Director, the Business Development Associate or the Project Managers. Questions or concerns from representatives of regulatory agencies are directed to the Laboratory Directors or a manager designated as a Deputy Laboratory Director. For any question or concern, the problem is first researched and the circumstances surrounding the incident are ascertained.

8.2 Where analytical results are at issue, the data is re-verified by the QA Manager or Department Manager. If the sample is still available a re-analysis may be requested to verify the original data. Where an error is identified, an amended report is issued.

8.3 The laboratory will gladly afford the client cooperation to clarify any client request to include laboratory visits and QC and PT data inspections.

8.4 If the investigation of any question raises doubt about compliance with the established laboratory policies, procedures or the quality of calibrations or test, the area of concern is audited in accordance with the audit section of this document. For more information on follow up documentation, refer to the Quality Control Follow-Up Forms SOP (Q24).

9.0 RECORD KEEPING

9.1 The laboratory information management system (LIMS) maintains records of Quality Control (QC) performance data and client samples data. All electronic and hard copy records are readily retrievable. For more information on data records, refer to the Records Management SOP (A02). It is the responsibility of the QA Manager or her designees to periodically review LIMS-generated records of QC performance data and to update acceptance criteria, as needed. For more information on the QC data review, refer to the Statistical Evaluation of Quality Control Data SOP (Q03).

9.2 Analytical run records are maintained as part of batch documentation for a particular analysis. An analytical run (listed in the LIMS as a “Batch”) can contain calibration standards, blanks, lab control samples (or blank spikes), replicate analyses, matrix

spikes, matrix spike duplicates, and sample data analyzed as a group and may contain one or more analytical batches. Raw data contains the following information:

9.2.1 Sample preparation.

9.2.2 pH observations including preservation confirmation (unless documented during splitting on work orders)

9.2.3 Sample matrix

9.2.4 Standard origin (See SOP Q05 section 4.0 for reagent origin.)

9.2.5 Method of analysis

9.2.6 Initials of analysts involved with the sample clearly indicating: "prepared by," "analyzed by" or "reviewed by".

9.2.7 Date and time of analysis.

9.2.8 Instrument i.d.

9.2.9 QC results and acceptance criteria.

9.3 All records are written in ink. Analysts are instructed to write legibly.

10.0 RECORDS MANAGEMENT

10.1 All hard copy and electronic records are stored on-site, or at an independent archive facility, for at least five years. After the archive period, the records are destroyed by a reputable records archiving and disposal company. Records maintained for at least five years include:

10.1.1 LIMS electronic files.

10.1.2 Raw analytical data files, hard copy and electronic.

10.1.3 QC data files including standard verification, control charts and corrective action reports.

10.1.4 Analytical data review reports.

10.1.5 Client files (hard copy and electronic) including all final reports, Chain of Custody forms, Analysis Request Forms, and other correspondence.

10.1.6 Personnel files including:

10.1.6.1 Personnel qualifications, experience, and training records.

10.1.6.2 Initial demonstration of capability (IDOC) and demonstration of continuing proficiency (DOCP) for each analyst or analytical work group.

10.1.6.3 A log of names, initials, and signatures for all individuals who are responsible for signing or initialing any laboratory records.

10.1.6.4 A record of Standard Operating Procedure (SOP) review and comprehension.

10.1.7 Business files including accounts payable, accounts receivable and payroll data.

10.2 Records are stored according to category in standard cardboard file boxes labeled with the category of records, date range, and descriptive information. A unique container number further identifies containers stored off-site. An access log is maintained for both on-site and off-site record retrieval.

10.3 There is a document control system indicating the time period during which a procedure, manual, or document is in effect. The steps of this process are outlined below. For more information regarding record management, refer to the Records Management SOP (A02) and the SOP Modification Policy (Q23).

10.3.1.1 Document Control Procedure

10.3.1.1.1 All documents issued to personnel, as part of the quality system, are reviewed and approved for use by authorized personnel prior to use. The QA Office is the issuing authority for all of the following documents.

10.3.1.1.2 Documents are uniquely identified by title and revision date.

10.3.1.1.3 Documents are reviewed and updated as needed by designated personnel who have access to pertinent background information.

10.3.1.1.4 Documents are protected electronically. All forms are stored as “read only”.

10.3.1.1.5 Only the most current version of each document is available for use.

10.3.1.1.6 A Master list of controlled documents is updated on an ongoing basis and located in QA Dept. on Nas-server/ESB SOPs

10.3.1.1.7 Outdated versions of quality documents are removed from circulation.

10.3.1.1.8 Obsolete documents retained for legal or knowledge preservation purposes are suitably marked to identify them as obsolete and saved on the Nas-Server or in hard copy form.

10.4 Laboratory management ensures that all requirements of the NELAP/ELAP and ISO Standard are in compliance where computers or automated equipment are used for the capturing, processing, reviewing, recording, reporting, storing or retrieving of test data.

10.4.1 Computer software programs and hardware utilized by the laboratory are installed, tested, updated and maintained by either the vendor supplying the software (such as instrumentation software) or by IS Concepts, Inc. an outside IT vendor. The documentation for all software, both commercial and custom written, as well as for all hardware is maintained in the storage area of the basement, by electronic log, or by IS Concepts. It is accessible during business hours. A detailed description of the lab's software, hardware, server and data security and retention protocol can be found in SOP Q17 "Good Automated Laboratory Practices (GALP)".

11.0 TEST METHODS UTILIZED BY THE LABORATORY

11.1 ESB uses appropriate test methods and procedures for all tests and related activities within its responsibility (including sample collection, sample handling, transport and storage, sample preparation, and sample analysis). The methods and procedures are consistent with the accuracy required, and with any standard specifications relevant to the calibrations or tests concerned.

11.1.1 The laboratory uses the most appropriate valid edition of a standard or method.

11.1.2 When the use of a specific test method for a sample analysis is mandated or requested, only the specified method is used.

11.1.3 When similar tests are combined in an analytical run, the most stringent method requirements are followed.

11.1.4 Where test methods are employed in a Performance Based Measurement System approach, the methods are fully documented and validated, and are available to the client and other recipients of the relevant reports.

11.2 Test Methods currently used by Babcock Laboratories may be found in *SW-846, Test Methods for Evaluating Solid Wastes Physical/Chemical Methods*, 3rd edition Update III 1996, *Methods for the Determination of Inorganic Substances in Environmental Samples*; EPA 600-R-93/100; *Methods for the Determination of Metals in Environmental Samples*; EPA 600-R-93/100; *Methods for the Determination of Metals in Environmental Samples, Supplement I*, EPA-600/R-95/111, EPA 500 and 600 series methods included or referenced in the Federal Register; and *Standard Methods for the Examination of Water and Wastewater*, 18th through 22nd editions, APHA/AWWA/WEF; Bacteriological Analytical Manual (BAM), 8th edition; Compendium of Methods of the Microbiological Examination of Foods (CMMEF), 4th edition; AOAC Official Methods of Analysis (OMA), 18th edition, or other approved or accepted methods.

12.0 HOUSEKEEPING AND SAFETY

12.1 Each analyst is responsible for keeping his or her work areas as neat and clean as possible. Each employee is warned of potential safety problems and is advised to be familiar with the following:

12.1.1 An eye wash station is available in each of the labs.

12.1.2 A drench-type safety shower is available in each of the prep labs and in the inorganic, microbiology, semi-volatile, and volatile labs.

12.1.3 Fire extinguishers are placed at several locations throughout the laboratory. They are easily found by signs that display their location. Analysts should be aware of the fire extinguishers located in their work areas. All fire extinguishers are serviced annually by contract with an outside company.

12.1.4 Chemical spill kits are centrally located and are available for solvents, mercury, caustics and acids.

12.1.5 First Aid kits are located in every lab. A fire blanket is located in each of the prep labs.

12.1.6 Flammable solvents are stored in an explosion proof cabinet with appropriate venting or below the hoods.

12.1.7 Evacuation plans are posted at several locations in the laboratory.

12.1.8 There is a Safety Data Sheet (SDS)/Material Safety Data Sheet (MSDS) Library centrally located and readily accessible to all personnel in the main hallway of 6100 Quail Valley Court.

12.2 Copies of the laboratory Emergency Plans are accessible to all personnel. Copies are located in the QA Office as well as on the NAS Server. These include a *Business Emergency Plan (BEP)*, *Chemical Hygiene Plan (CHP)*, *Fire Prevention Plan (FPP)*, and *Injury and Illness Prevention Plan (IIPP*).

13.0 SAMPLING (See Appendix F for Sampling Procedure References)

13.1 Each member of our Field Department has detailed knowledge of proper sampling techniques, sample handling procedures, and the criteria for sample acceptability. Field Department employees are trained in the proper safety requirements, heat illness prevention and precautionary measures to be used in field activities and have read and agreed to follow Field Sampling SOP (F02). The Field Department employees are advised to discuss with the on-site manager (if applicable) all steps necessary in obtaining the most representative sample possible – especially in unusual sampling situations. The Field Department and Log-in employees have each read and agreed to follow the Sample Acceptability SOP (A08). Samples are collected only in approved containers. Sample approval is based on the following requirements:

13.1.1 **Containers must be compatible with the sample and contain an adequate volume of sample.** The sample must not cause the container to corrode and the container must not contaminate the

sample. It must be of sufficient volume to hold enough sample for the required analyses – if not, multiple containers may be used, if needed.

13.1.2 **Containers must be made of approved materials.** For most uses, containers may be made of LPE plastic. Soda glass or borosilicate with Teflon is used for organic sampling. Plastic zip-lock bags or Mason jars are acceptable for many types of solid samples. For source gas emissions, tedlar bags, canisters, and absorbent traps are commonly used.

13.1.3 **Containers must be sterile for bacterial analysis.** Sterilized containers and lids are utilized for all bacterial analyses. A capacity of at least 100 ml is required. For bacteriological samples, we recommend sealing the containers in a zip-closure bag after collection for transport to the lab. Food and bottled beverage samples are submitted in their sealed product containers or bags as provided by the client.

13.2 The next section discusses other criteria used to determine sample acceptability including sample preservation, sample temperature, sample holding time, and condition of sample seal or evidence tape (if present).

13.3 The preservation and storage of samples varies according to the analyses to be performed. Proper preservation and storage may be found in *SW-846, Test Methods for Evaluating Solid Wastes Physical/Chemical Methods,* 3rd edition Update III 1996, in the *Handbook for Sampling and Sample Preservation of Water and Wastewater,* Sept 1982 EPA-600/4-82-029, 40CFR136 and 40CFR141.

13.4 For more information regarding sample collection, refer to the Field Sampling SOP (F02) or the UCMR3 Sampling SOP (F16). For sample containers, preservation and holding times refer to Appendix G – SOP (Q14).

14.0 SAMPLE IDENTIFICATION, CUSTODY, AND TRACKING

14.1 All written records are in indelible ink. For more information regarding sample acceptability, receiving and log-in, refer to the Sample Acceptability SOP (A08) and the Sample Receiving/Log-in SOP (A03).

14.2 Sample Identification

The samples are labeled in the field. The identification on the label includes the following. The information must be presented in such a way that the sample is uniquely identified and includes the following:

14.2.1 Identification of the sample.

14.2.2 The sampler's name.

14.2.3 The date the sample was taken.

14.2.4 The time (24-hour clock when possible) the sample was taken.

14.2.5 The client's name.

14.2.6 Significant information regarding the analysis (e.g. tests to be performed, temperature upon sampling, chlorine residual, preservation, if the sample is a composite or grab, miscellaneous comments, etc.)

14.3 Sample seals may be used to indicate possible tampering with the sample from the time of collection until the sample arrives at the laboratory – this is especially important for samples that may be used for litigation purposes and are delivered to the laboratory by the client or a third party.

14.4 Chain of Custody

14.4.1 A Chain of Custody form is filled out. A Chain of Custody is required for all samples and especially whenever the potential exists that the sample may be used for litigation. An example form is included in Appendix H. This form contains all of the above information included on the label and also "Relinquished by" and "Received by" blocks for the name(s) of the person(s) who submit or release the sample and the name(s) of the person(s) who receive the sample along with the date(s) and time(s) that the custody of the sample changes hands. The Chain of Custody also includes information relating to sample acceptability.

14.4.2 Information stored in LIMS serves as an internal Chain of Custody documenting sample use within the laboratory (section 14.7). If for litigation purposes a client requires the sample to be accessed only thru a custody officer, a form is available that tracks sample custody within the laboratory. When not in use the sample is kept in a locked refrigerator. For more information on litigation samples, refer to the Legal/Evidentiary Custody for Litigation Samples SOP (A01).

14.5 Sample Receipt Form

Whenever a Chain of Custody is not submitted with a sample, a Sample Receipt Form is filled out to document sample acceptability. This form lists the sample description and Babcock's laboratory number to unequivocally tie the form to the field sample. An example form is included in Appendix H.

14.6 Field Records

Field technicians maintain a logbook to document all of their sampling activities. This logbook contains pertinent information regarding collection of samples, including:

14.6.1 Name of contact

14.6.2 Location of sampling point

14.6.3 Date and time (24-hour clock) of collection

14.6.4 Field measurements

14.6.5 Comments

14.6.6 Any other information that is required by the Project Plan.

14.6.7 The field technician retains a copy of each Chain of Custody accompanying that day's samples.

14.7 Sample Tracking: Laboratory Information Management System

The laboratory has in place a LIMS for the tracking of all samples from the time they are logged in until the final report leaves the laboratory. At any step in the process, current information regarding the status of the sample can be obtained from the computer. The computer also keeps track of holding times and due dates.

14.8 Log-in of Samples

After the sample is collected, custody of the sample is turned over to the laboratory at the front counter. Log-in personnel verify sample acceptability – documenting acceptability on the Chain of Custody or a Sample Receipt Form. A Sample Acceptability checklist is kept in Log-in as a reminder of the criteria list. The client is contacted if any of the following occur:

14.8.1 The container is leaking or damaged.

14.8.2 The Chain of Custody seal is broken, if present.

14.8.3 The identification of the sample is not the same as that on the Chain of Custody (if the identification is not very different, the information is noted without necessarily contacting the client).

14.8.4 The sample is received past holding time (or likely to expire before the lab is able to perform the test).

14.8.5 The temperature of the sample exceeds method requirements.

14.8.6 The sterility of the sample container is questionable, if for bacteriological analysis.

14.9 The client must verbally authorize the lab to proceed with analysis if any of the above conditions are observed. This authorization is noted on the Sample Receipt Form or the Chain of Custody.

14.10 The Department Manager is informed if the samples are not preserved properly, incorrect containers are used, inappropriate sample size is provided, holding times have been exceeded, or any other problems occur so that corrective action, either in the lab or through the client, may be taken. Proper notation and warning is given before any sample is accepted under the conditions above.

14.11 Upon acceptance of the sample from the field technicians or directly from a client or courier, the sample is logged-in.

14.12 Every laboratory number or SampleID number that is assigned corresponds to a specific sample. The computer generates this number. The information in the computer unequivocally links the sample to the field identification. Other information recorded in the computer includes the following:

14.12.1 The name of the person, company, or agency requesting the analysis.

14.12.2 The sample description (corresponding to the field identification).

14.12.3 The date and time (24-hour clock) the sample was taken and the identity of the sampler.

14.12.4 The date and time (24-hour clock) the sample is submitted to the laboratory and the identity of the person submitting the sample.

14.12.5 The identity of the person logging in the sample.

14.12.6 The sample matrix.

14.12.7 The type of sample container.

14.12.8 Sample preservation (see also the Bottle Preservation SOP A09).

14.12.9 If evidence tape and/or seal are present.

14.12.10 Analyses requested - Constituents.

14.12.11 Chain of Custody (Y or N).

14.12.12 If thermal preservation is required, the temperature of the sample at receipt. (If recently collected, is the sample on ice?)

14.12.13 Any other pertinent information (such as any abnormalities or departures from the condition specified in the test method, reporting limit requests, high level QC or QC review requests, contact details, sample preservation exceptions).

14.13 After Log-in, two items are generated by the LIMS:

14.13.1 A Work Order Report containing information such as Client ID, Laboratory Number, analyses requested, and date/time of receipt is printed. Work Orders are kept sequentially in three-ring notebooks or file folders. Office staff verifies the log-in information and initials the work order. The Chain of Custody/Sample Receipt Form (including any common carrier documents received) and any other paperwork submitted with the samples are kept with the

Work Order Report. These documents are pulled for inclusion with the final report when the laboratory completes the analysis.

14.13.2 A durable, water resistant, computer-generated sample label is printed and affixed to each sample. Every sample container received from the client is uniquely identified on the label with the laboratory reference number and an alpha character (A, B, C) indicating the specific container. Sample preservation is verified and, if necessary, adjusted upon receipt or at the bench prior to analysis. For more information on sample preservation and storage, refer to the Sample Splitting, Preservation, Storage, and Disposal SOP (A06).

14.13.3 For microbiological samples, in addition to the Work Order Report and labels, the lab sheet may be printed from the LIMS.

15.0 IDENTIFICATION AND STORAGE OF SUBSAMPLES, EXTRACTS, AND DIGESTATES

15.1 Unique laboratory numbers generated by the LIMS identify all sample containers. When sub-samples, extracts and/or digestates are made, each additional container is uniquely identifiable. Sub-samples taken for preservation indicate the preservative added in addition to the SampleID number. Vials containing extracts for Organic determinations indicate the SampleID number, the Method Number and the extraction date. Digestate storage containers indicate the laboratory number and type of preparation – each preparation batch for trace metal analyses on any one date is also color-coded. All sub-samples, extracts, and digestates are stored according to the applicable preservation or the test method requirements.

16.0 TRANSPORTATION OF SAMPLES

16.1 Once samples have been received, they do not leave the premises. Office employees personally place the samples in the refrigerator (temp 2-6 °C), in the proper area of the sample storage room, or in the proper analytical section of the lab. Storage of samples must follow preservation protocols. Any relevant instructions regarding storage accompanying the sample must be followed. Samples are stored away from standards, food, and other sources of potential contamination. Any pertinent information regarding the samples must be discussed with the Department Manager.

16.2 All analysts (names and dates) handling the sample are documented electronically in LIMS or on the paperwork associated with the sample and its analysis. Samples that have been completed are held in the storage area for at least one month – at which time they are disposed of in the proper manner. For more information concerning sample disposal refer to Pollution Prevention SOP S07. Occasionally, samples for litigation purposes might be held for a longer period of time at the request of the client or samples may be returned to the client. All doors in the laboratory are locked securely and the entry alarm is armed when no approved personnel are present.

17.0 EMPLOYEE ORIENTATION AND TRAINING

17.1 All new employees to the company receive orientation to the company and its mission during new hire training and orientation. The orientation and training program is performed by Human Resources and the QA Department. Orientation includes familiarization with the Babcock Laboratories, Inc. Employee Handbook, the Quality Management Plan/Quality Assurance

Manual, the Ethics and Data Integrity Manual, and Laboratory Safety Training. Employees also receive training from the QA Department regarding QA procedures, NELAP/TNI/ELAP standards, ISO 17025 Standards (when applicable), laboratory techniques, LIMS, and company information. Employees are generally not released to their assigned department(s) until QA has completed their training. Training on specific job duties and responsibilities is performed by the Department Manager or an approved and trained designee. Specific job requirements such as knowledge, skill, accreditations, etc. are listed in the job descriptions included in attachment E. See Appendix I for examples of training module outlines.

17.2 The Employee Handbook discusses the standards of conduct, which are expected of all employees, including confidentiality of information. In detailing the contents of the Ethics and Data Integrity Manual, the employee also receives instruction regarding his/her ethical and legal responsibilities and the potential penalties for improper, unethical or illegal actions. All employees are required to sign a statement acknowledging they have been provided with this information and that they have read and are familiar with the manuals. Statements are on file in the QA Files located in the QA office and also on the computer in the QA folder on the NAS server. As stated in the Ethics and Data Integrity Manual (Appendix J), a Babcock Quality Control Corrective Action/Root Cause Analysis Report may be used to document challenges presented to the Ethics and Data Integrity Program. However, if any possibility of serious potential unethical behavior has been raised, an official Ethics Investigation will be initiated and the committee will report its findings to the CEO.

17.3 All employees receive refresher training for Ethics and Data Integrity, Quality Management Plan/QA Manual updates and

Laboratory Safety on an annual basis by the QA Department. Retraining or refresher training in all areas of the employee's job is performed on an on-going basis as needed. Employees are also provided the opportunity for continuing education through seminars, webinars, committee meetings and client training events (such as TEAM events) sponsored by the company.

18.0 STANDARD OPERATING PROCEDURES (SOPs)

18.1 Babcock maintains Standard Operating Procedures (SOPs) that accurately reflect all phases of current laboratory activities such as assessing data integrity, corrective actions, handling customer complaints, and all test methods. Some features of the SOPs include:

18.1.1 These documents are internally written documents.

18.1.2 For analytical tests, copies of published methods are used initially until the internally written SOP has been developed. Any deviations from the test method are documented *in italics*.

18.1.3 Copies of all SOPs are accessible to all personnel. Personnel must read all SOPs that are applicable to their assigned tasks and sign a statement that they have done so.

18.1.4 SOPs are organized.

18.1.5 Each SOP clearly indicates the effective date of the document, the revision number and the signature(s) of the approving authority (the Quality Assurance Manager or her designee).

18.1.6 Analytical SOPs are reviewed and updated during routine method audits. Non-analytical SOPs are reviewed and, where necessary, revised to ensure continuing suitability and compliance with laboratory policies and quality control standards

19.0 THE LABORATORY METHOD MANUAL

19.1 The laboratory has and maintains in-house Methods Manuals consisting of individual SOPs for each accredited analyte or test method. For more information regarding SOPs, refer to the SOP Modification Policy SOP (Q23). These manuals consist of copies of published or referenced test methods or the laboratory SOP. Each SOP includes or references, where applicable:

19.1.1 Identification of the test method.

19.1.2 Applicable matrix or matrices.

19.1.3 Reporting and Detection limit.

19.1.4 Scope and application, including components to be analyzed.

19.1.5 Summary of the test method.

19.1.6 Definitions (see also the Definitions SOP Q15).

19.1.7 Interferences.

19.1.8 Safety (see also the Safety SOPs S01-S07).

19.1.9 Equipment and supplies (see also the Equipment Maintenance SOP Q21).

19.1.10 Reagents and standards (see also the Reagent Quality and Documentation SOP Q05 and the Standard Quality and Documentation SOP Q08).

19.1.11 Sample collection, preservation, shipment and storage (see also the Bottle Control SOP Q13, the Holding Times/Due Dates SOP Q07, the Sample Containers, Preservation Techniques, and Holding Times for Aqueous Matrices SOP Q14 and the UCMR3 sampling and preservation guidelines SOP F16).

19.1.12 Quality control (see also the Quality Control Data SOP Q01).

19.1.13 Calibration and standardization.

19.1.14 Procedure (see also the General Laboratory Techniques SOP Q04 and the Good Automated Laboratory Practices SOP Q17).

19.1.15 Calculations.

19.1.16 Method performance.

19.1.17 Pollution prevention (see also Pollution Prevention SOP S07).

19.1.18 Data assessment and acceptance criteria for quality control measures (see also the Procedure for the Integration of Chromatographic Peaks SOP Q18).

19.1.19 Corrective actions for out-of-control data (see also the Corrective Action for Chemical Analyses SOP Q06).

19.1.20 Contingencies for handling out-of-control or unacceptable data.

19.1.21 Waste management (see also Pollution Prevention SOP S07).

19.1.22 References including instrument manual if applicable.

19.1.23 Any tables, diagrams, flowcharts and validation data.

19.2 It is the policy of Babcock Laboratories, Inc. that the Quality Control protocols specified by the Laboratory Methods Manuals be followed.

20.0 DEMONSTRATION OF PROFICIENCY

20.1 In order to demonstrate the suitability of a test method for its intended purpose, Babcock Laboratories, Inc. demonstrates and documents its ability to meet acceptance criteria either specified by the method, by the Environmental Protection Agency (EPA), the State program requirements or other accrediting bodies such as A2LA. Acceptance criteria meet or exceed these requirements and demonstrate that the test method provides correct/expected results with respect to specified detection capabilities, selectivity, and reproducibility.

20.2 Microbiological Analyses

The Microbiology laboratory utilizes accepted (official) test methods or commercialized test kits for official test methods. Proficiency with the test method is demonstrated prior to first use. Microbiological test methods are validated in terms of specificity and reproducibility by the use of positive and negative controls covering all aspects of the test. The validation of microbiological test methods is performed under the same conditions as those for routine sample analysis. Individual microbiologist demonstration of capability is required prior to unsupervised sample analysis. For details, please see the Bacteriology General Procedures and Quality Control SOP (B01).

20.3 Chemical Analyses

20.3.1 Laboratory

When initiating a new analytical method, the laboratory performs a complete data package including SOP, calibration, MDL study, and an Initial Demonstration of Capability (IDOC) as required by the method and/or the certifying agency. A follow-up is performed on any analyte that fails the laboratory acceptance criteria and an additional IDOC is performed for that analyte.

20.3.2 Work Cells

Whenever there is a significant change in the method or instrument, where Work Cells are used, the group must perform an Initial Demonstration of Capability (IDOC) as a unit. When a new analyst is added to the cell, the analyst may work with an experienced analyst until the training period is completed and four Laboratory Control Samples (LCSs) and Method Blank Samples (MBs) in four consecutive batches meet acceptance criteria. To demonstrate and document analyst proficiency, this data is recorded by the analyst using a Work Cell Change Form and submitted to the QA Department. If the new analyst cannot successfully complete the Work Cell Change, the cell must

perform a new IDOC or DOCP depending on the cell. In addition, if the entire Work Cell is changed/replaced, the Work Cell must perform an IDOC. (See instructions for IDOC in section 20.3.3.3 below).

20.3.3 Analyst Training Documentation

New employees or employees that are assigned new procedures undergo the following training:

20.3.3.1 The analyst reads the applicable SOP(s) and is shown the procedure by the Manager or an approved and designated trainer.

20.3.3.2 After the trainer has shown the new analyst the procedure once or more, as needed, the analyst performs the procedure (or their part of the procedure when working with a Work Cell or Work Group) under direct supervision of the trainer until the trainer is confident that the analyst can perform the procedure unsupervised.

20.3.3.3 The analyst performs an Initial Demonstration of Capability (IDOC). The IDOC is performed using either: 1) method specified procedure and criteria or, if the method does not specify 2) four aliquots of sample are analyzed at a concentration of 1-4 times the method or laboratory generated limit of quantitation. The aliquots may be either analyzed concurrently or over a period of several days (as long as they are consecutive for the analyst). The standards are from a source separate from the calibration. The average is calculated in the units used for reporting. The average and either the standard deviation or RSD are compared to method or, in the absence of method requirements, in-house acceptance criteria for recovery and reproducibility. If standard is not available for spiking, four sample aliquots are analyzed at a readable

concentration and compared to in-house acceptance criteria for reproducibility. After follow-up, test parameters that fail acceptance criteria are reanalyzed with additional aliquots until they meet the criteria. During this training period, the primary or secondary trainer will co-initial any analyses of client samples by the trainee. The completed IDOC certificate along with supporting raw data is kept in the QA files.

20.3.3.4 A training log is completed to document the training of the new analyst, including information such as the analyst name, trainer name, method, date SOP read, and date of IDOC or other demonstration.

20.3.3.5 The QA Department reviews proper documentation and calculations of the new analyst. The Demonstration of Capability Certificate Statement is approved by the Lab Director, and it is filed by the QA Department.

20.3.3.6 Annually, the analyst must perform a Demonstration of Continuing Proficiency (DOCP), as detailed in the Quality Control Data SOP (Q01). If the analyst has performed an IDOC during that calendar year, the IDOC will satisfy the requirement for that calendar year. If the analyst is part of a Work Cell that adds an employee with a Work Cell Change Form or has performed an IDOC those procedures will satisfy the requirement for that calendar year.

20.3.3.7 Demonstration of Capability Certification Statements -IDOC, DOCP, and Work Cell Change Form records are kept in the QA files.

Note: The training procedure is intended to ensure that personnel are adequately experienced in the duties they are expected to carry out and are

receiving any needed training. The Laboratory Directors or Lab Manager certifies that personnel with appropriate education and/or technical background perform all tests for which the laboratory is accredited. Employees are chosen for specific tasks based on their abilities and experience. Refer to A12 Employee Training Procedure SOP for more information.

21.0 METHOD DETECTION LIMIT

21.1 The **method detection limit (MDL) or Limit of Detection (LOD)** is verified statistically for each analytical method and for aqueous and non-aqueous matrices, as applicable. A minimum of seven replicates of a spiked matrix are processed and analyzed at a concentration of 2.5-5 times the estimated method detection limit or per method specifications. The standard deviation is calculated. The statistical method detection limit is the standard deviation multiplied by the student's T factor for the number of replicates at a 99% confidence level and multiplied by any preparation or dilution factor (the student's T factor for seven replicates at a 99% confidence level is 3.14). The reporting limit (or limit of quantitation (LOQ)) must be equal to or above the calculated statistical MDL except for special organic analyses (see Q01 for reporting limit requirements). MDL studies for each analytical procedure are performed per method requirements or whenever major changes in the instrument or procedure occur.

21.2 On an annual basis the laboratory must confirm the validity of either the LOD (for results reported as J flag values) or the LOQ.

21.3 Confirmation of LOD

21.3.1 The validity of the LOD is confirmed by qualitative identification of the analyte(s) in a QC sample in each quality system matrix containing the analyte at no more than 2-3X the LOD for the

single analyte tests and 1-4X the LOD for multiple analyte tests. This verification is performed on every instrument that is to be used for analysis of samples and reporting of data.

21.3.2 An LOD study is not required for any component for which spiking solutions or quality control samples are not available, or when test results are not to be reported to the LOD. Where an LOD study is not performed, the laboratory may not report a value below the Limit of Quantitation.

21.4 Confirmation of LOQ

21.4.1 The validity of the LOQ is confirmed by successful analysis of a QC sample containing the analyte(s) of concern in each quality system matrix 1-2 times the claimed LOQ. A successful analysis is one where the recovery of each analyte is within the established test method acceptance criteria or client data quality objectives for accuracy.

21.4.2 The LOQ study is not required for any component or property for which spiking solutions or quality control samples are not commercially available or otherwise inappropriate (e.g., pH) or if the LOD has already been confirmed.

22.0 PROFICIENCY TESTING (PT) SAMPLES

22.1 Babcock Laboratories, Inc. participates in Performance Testing (PT) studies at least twice each year in each field of accreditation (per matrix-technology/method-analyte/analyte group) in order to maintain accreditation. Certified samples are purchased, where available, from a National Voluntary Laboratory Accreditation Program (NVLAP) approved vendor. PT samples for microbiology food and bottled beverage testing methods such as pathogens are obtained from AOAC International. AOAC's PT program provides PT samples at a rate of four times per year. AOAC PTs for food and

bottled beverage analyses are completed at a minimum of two activities per method/test type and/or technology per year. The lab's entire scope is covered over a four-year period at minimum.

22.2 For environmental PT samples, if the laboratory receives a "Not Acceptable" result for a PT sample, a "Corrective Action: ESB PT Follow-up Form" is completed. The data is reviewed and the findings documented. These are reviewed by the QA Manager, signed by the Laboratory Director, and a copy is mailed to the accrediting authority/certifying body (for DMRQA studies, a copy is also sent to any affected DMRQA clients upon client request). If the laboratory receives a "Not Acceptable" result for two out of three of the last PT samples, the laboratory will order supplemental PT samples from an approved vender (such as Environmental Resource Associates or Absolute Standards) at least 15 calendar days apart from the closing date of one study to the shipment date of another study for the same field of proficiency testing, until a history of passing two out of three PT samples is reestablished. Proficiency testing raw data and reports are retained in accordance with the laboratory records management policy. For more information regarding our PT program, please refer to the Proficiency Testing SOPs (Q26 & Q27).

22.3 For food testing and bottled beverage PT samples, if the laboratory receives an "Unsatisfactory" of "Failed" result for a PT sample, the Root Cause of the unsatisfactory result is promptly investigated. The investigation is complete once the problem has been rectified and the lab is able to achieve satisfactory performances for the test/method in question.

23.0 QUALITY CONTROL OF LABORATORY CONDITIONS

23.1 For details on laboratory technique, equipment, and instrumentation refer to the Equipment Maintenance SOP (Q21) and the General Laboratory Technique SOP (Q04). The following is a general summary however, the current Q04 or Q21 SOP will supersede.

23.2 Equipment and Instrumentation

23.2.1 Examples of chemical instrumentation include Gas Chromatographs, Gas Chromatograph/Mass Spectrometers, Ion Chromatographs, Ion Chromatographs/ Mass Spectrometer/ Mass Spectrometers, High Performance Liquid Chromatograph, Inductively Coupled Plasma Emission Spectrophotometer, Inductively Coupled Plasma/Mass Spectrometer, Infra-red Spectrophotometer, UV-Visual Spectrophotometers, Total Organic Carbon Analyzer, nephelometers, recorders, and integrators. Bacteriological equipment includes incubators, autoclaves, fecal water baths, agar water baths, dry ovens, microscopes, stomachers and a Mini-Vidas. A list of current equipment, manufacturer, model, serial numbers, date received, date placed in service, condition when received, and laboratory location is kept on file. See Appendix K for equipment list.

23.2.2 All instruments/equipment are calibrated and maintained in accordance with manufacturer's specifications, method requirements, and well-established quality assurance practices. (See Appendix L: Calibration and Quality Control Criteria Charts for method specific calibration information.) A copy of the manufacturer's instructions, when available, is kept with the instrument. Equipment is operated by authorized personnel only. The analyst using the instrument/equipment maintains the

instrument in clean and operating order. Problems are reported immediately so that they can be corrected. When an instrument is taken out of use due to a maintenance problem, a sign is placed on the instrument indicating the instrument is out of service. All major instruments are kept on maintenance contracts. Maintenance logs are kept for all major analytical equipment. All maintenance procedures, routine and nonroutine, are clearly documented in the log. When a new instrument is placed into service it is first calibrated and checked to establish that it meets the laboratory's specification requirements and complies with the relevant standard specifications. MDL and IDOC studies are performed, and a “New Instrument Checklist” must be completed and approved by QA prior to sample analysis. For more information regarding equipment and a copy of the “New Instrument Checklist”, refer to the Equipment Maintenance SOP (Q21).

23.2.3 The temperatures of refrigerators, ovens, and incubators are monitored daily and recorded in a notebook along with the initials of the person performing the check. The temperatures of water baths are monitored daily (when in use). All thermometer calibrations are checked annually against an NIST certified thermometer at a level that is appropriate for its use. If a thermometer is broken, the calibration of the replacement is checked in such a way as to be NIST traceable before use. All calibration checks are recorded and any correction applied to the thermometer is recorded. A label indicating the calibration status is applied to each thermometer.

23.2.4 All glassware is rated as Class A. All glassware is cleaned to meet the sensitivity of the test method (e.g. acid rinsed for metal

determination or solvent rinsed for organic determination). See the appropriate analytical SOP for the glassware cleaning procedure required by that method.

23.2.5 Balances are calibrated and cleaned annually by an outside vendor. The sensitivity, reproducibility, and internal consistency are checked within the laboratory daily using certified weights. These weights are calibrated at least annually and the weight calibration certificates are kept on file in the QA office.

23.2.6 The pH meters are calibrated with each use using 7.0 and 10.0 buffers and verified by a 4.0 buffer.

23.2.7 The fume hood filter quality and velocities are monitored and recorded quarterly. Filters are changed when needed, typically on a quarterly basis. Fume hoods are serviced annually.

23.3 Environmental Conditions

23.3.1 The laboratory ensures that the environmental conditions do not invalidate results or adversely affect the required quality of any measurement. Measures have been implemented to isolate sensitive analyses and guard against cross contamination including restricting access to areas such as volatile analysis when solvent vapor contamination is a possibility. The laboratory adheres to specific environmental conditions specified in a test method or by regulation and where specified, documents compliance.

23.3.2 The laboratory monitors for contamination by processing a Method Blank with every analytical batch. In the event of contamination, environmental tests are stopped, and corrective

action is taken. The client is notified if it is believed the contamination has affected the client's sample result.

23.3.3 The laboratory takes into account factors that contribute to the total uncertainty of measurement in developing environmental test methods and procedures, in the training and qualification of personnel, and in the selection and calibration of the equipment it uses.

23.3.4 See section 24.2.2.4.4 or SOP Q03 "Statistical Evaluation of Quality Control Data" for details on uncertainty of measurement.

23.4 Reagents and Standards

23.4.1 Reagent water meets or exceeds ASTM Type II specifications. It is produced by a triple-stage commercial ion-exchange resin system. If higher quality water is desired, the water is then passed through a "Nanopure" system. All chemical reagents are ACS quality or better. Reagents are discarded and prepared fresh as required. The reagent name and date prepared shall serve as the unique identifier. Alternatively, some reagents are logged into Element and assigned a unique identifying number. If a second identical reagent is made on a given day, it will be designated with an additional letter so that the name and date remain unique. All standards are prepared from ACS reagents or purchased already standardized by a nationally known chemical manufacturer such as Baker, Eastman Kodak, B & J, Merk, Supelco, Mallinkrodt, Aldrich, Sigma, etc. The date received and opened is recorded on the bottle. New standard solutions are compared to a standard of a different manufacturer or lot number. If they fail to agree within method-acceptable criteria, then either the standard is re-made or both standards are compared to another standard from a third source.

23.4.2 Chemicals and laboratory supplies are purchased from approved vendors with an established history of reliability. A list of such vendors is maintained by the purchasing agent. Chemicals and supplies are selected by consulting approved methods for specific requirements that affect the quality of the environmental analyses. When purchasing a chemical or supply from a new supplier the purchasing agent or management identify deviations from the normal approved supplies and verify whether the new chemical is compliant with method requirements prior to purchase. Purchasing documents for items affecting the quality of laboratory output contain data describing the services and supplies ordered. These documents are reviewed. Before starting a new analytical procedure, the responsible persons review the chemical types to be used. For more information regarding purchasing and suppliers please see SOP A07 “Chemical Procurement Distribution and Storage”.

23.4.3 Standard and reagent logs are maintained either in a logbook or LIMS to document the traceability of standards and reagents and to record the manufacturer, lot number, concentration, preparer, and date of preparation. Whenever a new product is ordered, it is checked to verify that it meets specific method requirements.

23.4.4 For further details on the handling and use of reagents and standards, refer to the Reagent Quality and Documentation SOP and the Standard Quality and Documentation SOP (Q05 and Q08, respectively).

24.0 QUALITY CONTROL OF ANALYTICAL PROCEDURES

24.1 See the Quality Control Data SOP (Q01) and Bacteriology General Procedures and Quality Control (B01) for details and Appendix M for QA References.

24.2 Chemical Determinations:

24.2.1 Method QC

Calibration curves and linearity checks are run as prescribed in the applicable method for each procedure and for all parameters. All quality control requirements of each method must be met.

24.2.2 Batch QC

24.2.2.1 A batch is a set of 20 or fewer samples of a similar matrix that are processed together with the same method and personnel, using the same lots of reagents.

24.2.2.2 A Laboratory Control Sample (LCS) or Blank Spike (BS), a Method Blank (MB), a Matrix Spike (MS) and Matrix Spike Duplicate (MSD), sample Duplicate (Dup), or Laboratory Control Sample Duplicate (LCSD) may be analyzed for each batch per method requirements. A Dup is analyzed in place of the MS and MSD for gravimetric or titrimetric analyses. Where the method does not require MS/MSD or Dup, an LCS duplicate (LCSD) may be used to gather precision data. Note: Other documents may refer to these QC samples in different ways. For example, EPA 500 series methods refer to an LCS as a Laboratory Fortified Blank (LFB) and an MS as a Laboratory Fortified Matrix (LFM). Element refers to an LCS as a Blank Spike (BS).

24.2.2.3 Batch Controls:

24.2.2.3.1 The MB is used to assess the preparation batch for possible contamination during the preparation and processing steps. The MB shall be processed along with and under the same conditions as the associated samples to include all steps of the analytical procedure. The MB documents the purity of any reagents or waters used. Under NELAP/ELAP/ISO rules, a MB shall consist of a matrix that is similar to the associated samples and is known to be free of the analyte of interest.

24.2.2.3.2 The LCS is used to evaluate the performance of the total analytical system, including all preparation and analysis steps. The LCS is analyzed at a minimum of 1 per preparation batch. Exceptions would be for those analytes for which no spiking solutions are available. The LCS is a controlled matrix, known to be free of analytes of interest, spiked with known and verified concentrations of analytes.

24.2.2.4 Sample Specific Controls

24.2.2.4.1 Sample specific controls determine the effect of the sample matrix on method performance. They are designed as data quality indicators for a specific sample using the designated test method. These controls do not evaluate laboratory performance of an entire batch. Examples include: MS, MSD, Dup and surrogate spikes (Surr).

24.2.2.4.2 MS/MSD indicates the effect of the sample matrix on the precision and accuracy of the results generated using the selected method. The information is sample/matrix specific and would not normally be used to determine the validity of the entire batch.

24.2.2.4.3 Duplicates are replicate aliquots of the same actual sample taken through the entire analytical procedure to indicate the precision of the results for the specific sample using the selected method. They provide a usable measure of precision only when the target analytes are found in sufficient quantity in the sample chosen for duplication.

24.2.2.4.4 The evaluated MS recoveries (or duplicate data in the absence of MS data) are in our estimation a valid approximation of method uncertainty for that matrix. MS data is an indication of correctness and reliability of the environmental tests including contributions from: human factors, accommodation and environmental conditions, environmental test methods and method validation, equipment, measurement traceability, sampling, and the handling of samples. MS and duplicate recoveries are presented to the user in a standard report. See SOP Q03 section 5.0 for more information.

24.2.2.4.5 For some analyses such as microbiological analyses certified under ISO 17025 requirements an alternative procedure for estimating the uncertainty of measurement is utilized. This procedure compiles all the factors listed above and statistically derives a calculated measurement of uncertainty through the use of a software program provided by an outside vendor such as Calibration Laboratory Assessment Program (CLAS). See SOP Q03 section 6.0 for more information.

24.2.2.4.6 Surrogate compounds represent the various chemistries of the target analytes in the method. They are deliberately chosen because they are unlikely to occur as an environmental contaminant.

24.2.2.5 Other QC

24.2.2.5.1 External QC reference samples are obtained from an outside source for inclusion in our procedures for method verification. External QC reference samples from an appropriate source, such as Environmental Resource Associates or Absolute Standards, are analyzed semi-annually.

24.2.2.5.2 Internal standards are routinely included with determinations of metals and organics; also methods of addition may be incorporated. An internal standard (IS) is a pure compound that is not a contaminant in the sample and is added to a sample or sample extract in a known amount.

The IS is used to measure the relative responses of target analytes and surrogates that are in the same sample. When an IS is used, it is added to the samples and QC samples or their extracts.

24.2.2.5.3 Non-routine samples with complex or unfamiliar matrices might need special QC, such as additional procedural spikes or two or more dissimilar methods of analysis. All batch QC is reviewed daily.

24.3 Bacteriological Determinations:

There are general procedures and quality control (QC) requirements that are unique to Bacteriological Determinations. These include sample container sterility checks, glassware cleaning procedures, housekeeping requirements, media maintenance/preparation/QC, dilution water requirements, instrument calibration, monitoring of incubators/water bath/dry oven, autoclave use and QC documentation (including the use of biological indicators of sterilization efficiency), reference culture requirements, and quality control analyses. For details, please refer to the Bacteriology General Procedures and Quality Control SOP (B01).

25.0 STATISTICAL EVALUATION OF DATA FROM CHEMICAL ANALYSES

25.1 The laboratory has quality control procedures for monitoring the validity of analytical tests undertaken. The resulting data is recorded in such a way that trends are detectable (refer to the Statistical Evaluation of Data SOP (Q03)), and when practical, statistical techniques are applied to the reviewing of the results (please refer to section 26).

25.2 All samples are analyzed within analytical batches. An analytical batch includes the QC samples discussed in Section 24 and any calibration and instrument checks required by the method.

25.3 Calibration curves for most analyses are a minimum of three points. Some methods require additional points in the calibration curve. Calibration requirements that are specified in the applicable test method must be met. See Appendix L for Calibration and Quality Control Criteria Charts.

25.4 The data from LCSs, MSs, MSDs, Duplicates and Surrogates are used for statistical evaluation. This is based on the following examinations:

25.4.1 From duplicates, both of spikes and samples, precision data is calculated and the **Relative Percent Difference (RPD)** is determined. The equation is:

$$\text{Relative Percent Difference} = \frac{|A - B|}{\left(\frac{A + B}{2}\right)} \times 100$$

Where *A* is the analytical result for the matrix spike (or sample) and *B* is the analytical result for the matrix spike duplicate (or sample duplicate).

25.4.2 From results of the **MS/MSD** (S) accuracy data is calculated and the **percent recovery (%Rec)** is determined. The equation is:

$$\text{Percent Recovery} = \frac{|\text{M - A}|}{\text{Q}} \times 100$$

Where M is the matrix spike analytical result, A is the analytical result of the (unspiked) sample, and Q is the amount of spike added.

25.4.3 An **LCS percent recovery (%Rec)** is calculated by comparing the LCS analytical result (A) to the "True" value, which is the expected value of the spike (Q) (or the historical average of the control).

$$\text{Percent recovery} = \frac{A}{Q} \times 100$$

If the percent recovery is not within the laboratory acceptance criteria, the analysis is considered to be “out of control” and will not continue until the cause is found and corrective measures are taken. Any affected samples associated with the out-of-control LCS are reprocessed for re-analysis or the results are reported with appropriate data qualifying codes.

25.4.4 The **relative standard deviation (RSD)** may be determined from the QC data using the following equation:

$$\text{Relative Standard Deviation} = \frac{S}{\overline{X}} \times 100$$

Where:

$$S = \sqrt{\frac{\sum (X - \overline{X})^2}{N - 1}}$$

Where S is the standard deviation, $\overline{X}$ is the mean of the observed value, X is the observed value, and *N* is the number of observations.

25.5 Control charts are generated in the LIMS. For details, please refer to the Statistical Evaluation of Data SOP (Q03).

26.0 DATA REDUCTION AND VALIDATION

26.1 To ensure the quality of the data, several review steps are incorporated into the data review process. For details refer to the

Data Review and Validation SOP (Q10). In summary, the first level of review is the analyst who preliminarily assesses whether the batch QC acceptance criteria are met, adds qualifiers as appropriate, checks historical data (if available) and checks calculations, units, significant figures and dilution or concentration factors. The Peer Reviewers performs the second level of review. The Final department review follows, including a check to determine if proper relationships exist among the parameters in the sample. This review step is performed by the manager or approved designee. The Standard Methods SM 1030 E procedure may be also used along with the following:

26.2 Mineral Balances

Equation (1) for Total Cations (me/L)

Ca + Mg + Na + K + NH_3-N = total cations

Equation (2) for Total Anions (me/L)

CO_3 + HCO_3 + OH + SO_4 + Cl + NO_3 + F = total anions

Equation (3) for Calculated TDS (mg/L)

Calculated TDS = Ca + Mg + Na + K + Cl + SO_4 + NO_3 + F + SiO_2
+ (0.6 $\times$ Total Alkalinity as $CaCO_3$)

The measured TDS may be higher than the calculated TDS because a significant contributor may not be in the calculation.

Equation (4) for Cation/Anion Balance

$$\text{Balance Acceptance Criteria} = 100 \times \frac{(\text{total cations - total anions})}{(\text{total cations + total anions})}$$

The result should be 100 $\pm$ 5%.

Equation (5) for Calculated Specific Conductance

EC (Calc) = (total cations + total anions) $\times$ 50

(Both the total anion and total cations should be 1/100 of the measured EC value.) The calculated EC is expected to be within 10% of the measured EC. Thus:

Equation (6)

$$0.9 \leq \frac{EC_{calculated}}{EC_{measured}} \leq 1.1$$

If the ratio of TDS to conductivity falls below 0.55, the lower ion sum is suspect and reanalyzed. If the ratio is above 0.7, the higher ion sum is suspect and reanalyzed. If reanalysis causes no change in the lower ion sums, an unmeasured constituent, such as nitrite or organic acids may be present at significant levels. If poorly disassociated calcium and sulfate ions are present, the TDS may be higher than the EC. The acceptance criterion is as follows:

Equation (7)

$$\frac{TDS_{measured}}{EC_{measured}} = 0.55 - 0.70$$

and/or

$$\frac{TDS_{calculated}}{EC_{calculated}} = 0.55 - 0.70$$

Reference: Standard Methods for the Examination of Water and Wastewater, APHA, AWWA, WEF, 22nd edition

26.3 Demand Ratios

A general rule of thumb is:

BOD = 0.40-0.60 of COD

TOC = 0.40 of COD (approximately)

TOC = 0.60 of BOD (approximately)

26.4 Nutrient Relationships

Total Nitrogen = Organic Nitrogen + Inorganic Nitrogen

Inorganic Nitrogen = NO_3-N + NO_2-N + NH_3-N

Kjeldahl Nitrogen = Organic Nitrogen + NH_3-N

Organic Nitrogen = Kjeldahl Nitrogen - NH_3-N

The above nitrogen relationships are checked to ensure proper calculations have been performed.

26.5 Trace Organic Contaminants:

TOX = Volatile Organic Halogens + Non-volatile Organic Halogens

Volatile Organic Halogens = Polar + Non-polar Volatile Organic Halogens

Non-volatile Organic Halogens = Polar + Non-polar Non-volatile Organic Halogens

Non-polar Volatile Organic Halogens are measured from EPA Methods 524, 624, or 8260.

Non-polar Non-volatile Organic Halogens are measured from EPA Method 525, 625, or 8270 analysis.

From the above relationships, the following is performed:

TOX $\geq$ Non-polar (Volatile + Non-volatile) Organic Halides

26.6 The Department Managers or Lab Director approves the results in the computer when all analyses requested on the sample are completed.

27.0 FINAL REPORT REVIEW

27.1 For some reports the QA Manager or Project Manager (or other authorized person) will also review the data just prior to generating the final report. These reports are generally given an extra review due to project-specific requirements such as an analyte list or reporting limit that is not typical, such as a request to report of J-flag values, or a request for a higher level QC (see Section 29 for further details on reporting of results). The initials and date of review are recorded on the Work Order report or recorded in LIMS as part of the data audit trail.

27.2 After the final report is generated, the Project Managers (or other authorized signatory) will be responsible for final review and signing of the report. Electronic Signatures are utilized for reports that are delivered electronically to clients.

27.3 Each reviewer (Analyst, Peer Reviewer, Manager, QA Manager/Project Manager, and Laboratory Director) verifies that the data have been reported accurately, clearly, unambiguously, and objectively.

28.0 CORRECTIVE ACTION FOR OUT-OF-CONTROL QC

28.1 For details on corrective action, see the Corrective Action SOP (Q06) and the SOP for the method. The following is a general summary however, the current Q06 or method SOP will supersede.

28.2 For Chemical Determinations

28.2.1 Corrective action is necessary when the upper or lower control limits for the test parameters have been exceeded for laboratory control samples or when processed blanks show an unacceptable level of contamination.

28.2.2 The first step taken when QC results are "out of control" is to recheck all mathematical calculations including such items as concentration and/or dilution factors and calibration curve readings.

28.2.3 If the first step fails to solve the problem then the reagents are checked for proper chemical reaction, for example, the esterification potency for EPA Method 8151.

28.2.4 Reagents and glassware are checked for contamination. Reagent blanks are checked containing the acids used in metal digestion or the solvents used in organic analysis.

28.2.5 Standards are checked for proper concentration. New standards from a different supplier/lot number are prepared to check against the standards used during the analysis in question.

28.2.6 When a batch of data is transferred or manually entered into the laboratory database and later found to be in error OR the need for re-running for verification, a follow-up is initiated by the analyst, with corroboration from the Peer Reviewer and Manager. The answers in the database are flagged as suspect until the problem can be resolved and the status remains at pending. Samples affected are either reanalyzed or verified. If the problem is not corrected, or the holding time has been exceeded or there is insufficient sample for a follow-up—the client is notified so that it can be determined if the site should be resampled. If suspect results are reported, the report is flagged with a note indicating the problem.

28.3 For Bacteriological Determinations

28.3.1 Corrective action is necessary when QC cultures show atypical response or when check sample results exceed the given acceptance criteria. The associated samples and quality control data are reviewed to verify sterility and findings are documented on the associated lab sheet(s), in LIMS or a follow-up form.

28.4 For All Analyses

28.4.1 Quality Control Non-Conformance Forms (Formerly identified as ESB Analysis dispute and QC Follow-Up Form) are generated when QC problems cannot be corrected during the run and documented in the analytical data itself. These forms are utilized to document the situation, corrective actions taken and final resolution. Refer to SOP (Q24) for further details. As appropriate for the test method in question, further corrective actions, such as troubleshooting instrumentation and re-calibration may be performed.

28.4.2 Quality control data is analyzed for all analyses. When this quality control data is found to be outside pre-defined criteria, planned action is taken to correct the problem and to prevent the reporting of incorrect results.

28.4.3 Any trends noted regarding batch QC failures, analytical issues or non-conformances are subject to a Root Cause Analysis Investigation in order to correct the issue and to also prevent any reoccurrences. Areas for Continuous Improvement are often identified during Root Cause Analysis Investigations as well.

29.0 REPORTING OF RESULTS

29.1 Analytical Chemistry Reporting Procedure

The final copy of the report contains all information necessary for the interpretation of the test results and all information required by the method user presented in a simplified and straight forward manner– including the following information:

29.1.1 A report title.

29.1.2 The name of the laboratory.

29.1.3 The address of the laboratory.

29.1.4 The phone number of the laboratory.

29.1.5 Laboratory identification number.

29.1.6 Unique page identification.

29.1.7 Name and address of client and project name, if applicable.

29.1.8 Description and unambiguous identification of test sample—including the client identification code.

29.1.9 Analytical results including units and reporting limit (RDL).

29.1.10 Identification of any quality control failure within the batch that might affect the validity of the result by use of appropriate data qualifiers.

29.1.11 The reported units for samples are as received, unless identified as “dry” (corrected for dry weight).

29.1.12 Qualification of results derived from samples that did not meet acceptance requirements – such as improper container, holding time or preservative.

29.1.13 Date of receipt of sample, name of the submitter, date and time of sample collection, name of the sampler (if known), date(s) and time(s) of analytical test(s), and analyst(s) initials.

29.1.14 Identification of the test method used or a description of any non-standard test used.

29.1.15 Any other information relevant to the specific test.

29.1.16 Definition of data qualifiers.

29.1.17 The approval signature and title of the signatory (or electronic equivalent).

29.1.18 A statement attesting to the quality standards that are applied to the generation of analytical results or where applicable, reasons that the analysis did not conform to required specifications.

29.1.19 Where applicable, clear indication of numerical results with values outside of quantitation limits.

29.1.20 Where applicable, a statement to the effect that the results relate only to the items tested or to the sample as received by the laboratory.

29.1.21 Where relevant, a statement that the certificate or report shall not be reproduced except in full, without the written approval of the laboratory.

29.1.22 Where applicable, clear identification of all data provided by outside sources, such as subcontracted laboratories, clients, etc.

29.1.23 Where applicable, amended reports include a case narrative or cover letter indicating that they are amended, the reason for amendment, and the date of the previous report that they supersede.

29.1.24 Where applicable, Non- accredited work is clearly identified by including a note in the analyte name, or by a "*" in the units of the affected analyte, along with a definition footer in the report.

29.1.25 Where applicable, the laboratory provides all the required information to their client for use in preparing monthly regulatory reports.

Note: A summary report is available upon request. If some of the above information is not presented in this report format it is readily available from the laboratory.

29.2 Reporting Records

All paperwork that is submitted by the client and the original Chain of Custody forms are attached to the work order. This is stored in the client files. Whenever format allows, electronic copies of amended reports are traceable internally by a date and time stamp.

29.3 Subcontracted Analyses

29.3.1 The laboratory clearly indicates to the client its intention to subcontract laboratory work in its contract bids and bid quotes.

29.3.2 When a subcontract lab is utilized, clients are sent the original reports from the subcontract lab. Babcock Laboratories, Inc. does not report subcontracted results on company letterhead (with the exception of transcribed results onto state forms for reporting to the state Office of Drinking Water. This is clearly noted on the report.)

29.3.3 All subcontracted analyses are to be performed by a NELAP/ELAP accredited laboratory or by a laboratory that meets applicable statutory and regulatory requirements for performing the tests. Subcontract laboratories' current certifications are kept on file in the office of the Regional Business Development Director.

29.3.4 Subcontract laboratories are audited periodically. Audits consist of a two to three person panel over a 4 hour time period, reviewing laboratory documents, touring the facility, and questioning staff to determine if Babcock quality standards and those required for accreditation are being upheld.

29.3.5 Subcontract results are reviewed prior to sending to the client. Verification of analysis reporting levels in the form of a calibration standard at the RL is kept on file.

29.3.6 For more information refer to the Review of Contracts and Tenders SOP (A13).

29.4 Mandated Verbal Notification to Water Systems

29.4.1 The laboratory notifies the water system personnel immediately if Coliform is found in the presence/absence Coliform test, there are any Coliform positive tubes, or the sample is declared invalid due to a turbid culture with the absence of gas production using either the multiple tube fermentation technique or the presence/absence Coliform test. Following the Total Coliform Rule, follow-up samples are taken until the samples are negative for Coliform (see the Microbiology Notification and Reporting Procedures SOP BO8, for further details).

29.4.2 The laboratory notifies the water system personnel if a final drinking water nitrate result exceeds the MCL of 45 mg/L (as NO_3, or 10 mg/L as N). Notification is performed within 24 hours of obtaining a verified result. Once notified, the client must resample and the laboratory must reanalyze within a 24 hour period. Refer to Inorganic Ions by Ion Chromatography SOP I19 for further details.

29.4.3 The laboratory notifies the water system personnel if a final drinking water perchlorate result exceeds the MCL of 6ug/L. Notification is performed within 48 hours of obtaining a verified result. Once notified, the client must resample and the laboratory

must reanalyze within a 48 hour period. Refer to Perchlorate by Ion Chromatography SOP I19B for further details.

29.5 Quality Control Reports:

29.5.1 QC data is available for all chemical batches and are reported to the client upon request. Each project will be assigned a type of data package (or QC Level) based on the objectives of their project and this will determine the amount of QC data included in the final report.

29.5.2 The **Level I or "short report**" data packages are created from data in the Laboratory Information Management System (LIMS, Element). Level I data packages receive our general data review procedure and include Client Information, Work Order, Sample Information, Analyte(s), Result, Reportable Detection Limit (RDL), Units, Method, Analysis Date, and Analyst information. Data qualifier flags will only appear as needed.

29.5.3 The **Level II or "standard report"** data packages are created from data in the LIMS. Level II data packages receive our general data review procedures and review by a Project Manager or QA Manager (the Work Order Report will indicate that the report "Needs QC"). Standard reports include all elements of the short report. In addition, the Batch Quality Control data for the QC samples are provided. The Batch ID and Method appear as the heading above each set of Batch QC. Each QC sample will have information on the Date Prepared, Date Analyzed, Analyte(s), Result, Reportable Detection Limit (RDL), and Units. As discussed in Section 24, the QC samples will vary by method but LIMS reports may include data on the Blanks, Laboratory Control Samples/Spikes, Laboratory Control Samples/Spikes Duplicates,

Matrix Spikes, Matrix Spike Duplicates, and Sample Duplicates. Where applicable, the following data are included with each type of QC sample:

29.5.3.1 Laboratory Control Samples/Spikes:

29.5.3.1.1 Spike Level and

29.5.3.1.2 Accuracy (Percent Recovery [%Rec] and %Rec Limits)

29.5.3.2 Laboratory Control Samples/Spikes Duplicates:

29.5.3.2.1 Spike Level

29.5.3.2.2 Accuracy (%Rec and %Rec Limits), and

29.5.3.2.3 Precision (Relative Percent Difference [RPD] and RPD Limit)

29.5.3.3 Matrix Spikes:

29.5.3.3.1 Source Result,

29.5.3.3.2 Spike Level, and

29.5.3.3.3 Accuracy (%Rec and %Rec Limits)

29.5.3.4 Matrix Spike Duplicates:

29.5.3.4.1 Source Result,

29.5.3.4.2 Spike Level,

29.5.3.4.3 Accuracy (%Rec and %Rec Limits), and

29.5.3.4.4 Precision (RPD and RPD Limit)

29.5.3.5 Sample Duplicates:

29.5.3.5.1 Source Result and

29.5.3.5.2 Precision (RPD and RPD Limit)

29.6 Higher level data packages are created from data in the LIMS and also include special data packages created by a Project Manager. Higher level data packages receive our general data review procedures and review by a Project Manager. **Level III** data packages include all elements of the standard report with the addition of run logs/bench sheets and calibration curves. **Level III+**

data packages include all elements of the standard report with the addition of run logs/bench sheets, calibration curves, and raw data (chromatograms etc.). **Level IV** data packages include all elements of the standard report with the addition of run log/bench sheet, calibration curves, raw data (chromatograms etc.), and standard logs. **Custom** QC packages, electronic versions of the data, and other variations are also available to meet the specific needs of each project and will be established on client/project basis.

29.7 In addition, reports are available with **J-flag** data. J-flag reports include estimated values for results that fall between the Method Detection Limit (MDL) and Reportable Detection Limit (RDL). The MDL is listed for each analyte. A J-flag report receives our general data review procedures and a review by a Project Manager or QA Manager (the Work Order Report will indicate that the report needs "J-flag").

30.0 AUDITS

30.1 Method Audits

Audits of all analytical methods are performed periodically and in accordance with a predetermined schedule, conducted by the Quality Assurance Department to ensure that its operations continue to comply with quality assurance policies, the requirements of the management system, method and NELAP, ELAP or ISO 17025 requirements. QA Department personnel are independent of the activities being audited (See Appendix N for example Audit Forms).

30.2 Internal Quality System Audits

The QA Manager and/or other trained and qualified personnel conduct an annual audit of the laboratory following the

NELAP/ELAP and ISO 17025:2005 requirements to determine that the laboratory operation continues to comply with the laboratory's quality and management systems. Quality System Checklist such as the A2LA "C204" Specific Checklist - Combined ISO-IEC 17025 and Food and Pharmaceutical Testing Laboratory Accreditation Program Requirements are used to conduct this audit. Laboratory documentation used to satisfy these requirements are referenced on the checklists. Results of these audits are reported to management for review. If results cast doubt on the correctness or validity of the laboratories calibrations or test results, the laboratory shall take immediate corrective action and shall immediately notify, in writing, any client whose samples were involved. For more information regarding these audits, please refer to the Audit Standard Operating Procedure (Q16).

30.3 Managerial Audits

The QA Manager annually conducts a review of the quality plan and management system as well as its testing and calibration activities. The purpose of this annual review is to ensure the quality plan and management's systems continuing suitability and effectiveness and to introduce any necessary changes or continuous improvements in the effectiveness of the quality system, management system and laboratory operations. The review addresses the suitability of policies and procedures, reports from managerial personnel, the outcome of recent internal audits, assessments by external bodies, the results of interlaboratory comparisons or proficiency tests, any changes in the volume and type of work undertaken, feedback and complaints from clients, corrective and preventive actions, outcome of Root Cause Analysis investigations including recommendations for continuous improvement and other relevant factors such as Quality Control activities, resources, and staff training. The

corrective and preventative actions section include actions taken concerning necessary improvements mentioned in the previous year's managerial and quality system audits. The laboratory has a procedure for review by management and maintains records of review findings and actions. For more information regarding these audits, please refer to the Audit Standard Operating Procedure (Q16). Annual Managerial Audit reports are kept on file in the QA office, the Nas Server and available upon request.

30.4 Audit Reviews

All audit and review findings and any corrective actions that arise from them are documented. Findings are reviewed with management and staff in order to identify and implement corrective actions, process improvement opportunities, and to ensure continuous quality improvement. The laboratory management ensures that these actions are discharged within the agreed time frame.

31.0 Quality Management Plan/Quality Assurance Manual – Updates and Reviews:

31.1 This QMP/QA Manual is designed to be compliant with NELAP/ELAP requirements ,ISO 17025 requirements as well as EPA document QA/R-2. The contents of this manual are reviewed annually (at a minimum) by the QA Manager and the Laboratory Directors for compliance to applicable regulations – ensuring that it reflects existing practices within the laboratory.

31.2 The QA Manual clearly indicates the revision month and the effective date. The effective date indicates that at midnight at the start of the effective date the new manual goes into effect.

31.3 All signatory personnel within the laboratory document their approval by signing of each new version of this document prior to its release.

31.4 The QA Manager maintains an archive of previous versions of this document.

31.5 All employees read the effective revision of the QMP/QA Manual and are familiar with its contents. Each employee signs a statement that he/she has read the QA Manual and declares his/her intention of complying with the requirements contained within Babcock Laboratories, Inc. QA Manual.

32.0 **APPENDICES** (Available Upon Request)

A. Copies of Certifications

B. R105-Requirements When Making Reference to A2LA Accredited Status***

C. Resumes of Key Personnel

D. Organizational Chart

E. Job Descriptions of Key Personnel

F. References for Sampling Procedures***

G. Sample Preservation and Holding Times***

H. Sample Forms: Chain of Custody and Sample Receipt

I. Training Module Outlines***

J. Ethics and Data Integrity Manual

K. Equipment List

L. Calibration and Quality Control Criteria Charts***

M. References for QA Procedures

N. Documentation Audit Forms

O. Sample Transportation for Third Party Couriers (SOP F14)***

***Note: Noted appendices and referenced SOPs are available upon request.

Appendix A Copies of Certifications

Appendix B R105-Requirements When Making Reference to A2LA Accredited Status **(Available Upon Request)**

Appendix C Resumes of Key Personnel

Appendix D Organizational Chart

Appendix E Job Descriptions of Key Personnel

Appendix F References for Sampling Procedures (**Available Upon Request)**

Appendix G Sample Preservation and Holding Times (**Available Upon Request**)

Appendix H Sample Forms:

Chain of Custody and Sample Receipt

Appendix I Training Module Outlines (Available Upon Request)

Appendix J Ethics and Data Integrity Manual

Appendix K Equipment List

Appendix L Calibration and Quality Control Criteria Charts (**Available Upon Request)**

Appendix M References for QA Procedures

Appendix N Documentation Audit Forms

Appendix O Sample Transportation for Third Party Couriers (SOP F14)
(Available Upon Request)

Resumes of Key Personnel - available upon request

Lawrence Chrystal
Allison Mackenzie
Paul Monroy
Tiffany Gomez
Cathleen Iijima
Cyndi Moore
Mark Tracy
Stacey Fry
Carol Kase
Susann Thomas
Tiffany Hudson
Aurea Yogarajah
Fred Kent
William Bayle
Julia Sudds
LuAnn Thomas
James Schaupp
Caroline Sangari
Salwa Loucka
Enrique Aguilar
Evelyn Calderon
David Schiessel

Note: This is the last page of the QA Manual (version effective September 30, 2014)

BIOCHEMICAL OXYGEN DEMAND - 5 DAY TEST
Babcock Laboratories, Inc.
STANDARD OPERATING PROCEDURE
(**Standard Methods 5210 B**)
Date Effective: 04/07/14

1.0 Scope and Application: This method is used to determine the relative oxygen requirements of wastewaters. This method may also be used for other matrices if both client and regulator agree to this variation.

2.0 Reporting limits: The reporting limit for this method is 5.0 mg/L for a 40% dilution. Refer to Element for MDL information.

3.0 Summary of Method: The method consists of placing a sample in a full airtight bottle and incubating the bottle under specified conditions for a specific time. Dissolved oxygen is measured initially and after incubation. The BOD is computed from the difference between the initial and final dissolved oxygen.

4.0 Environmental Relevance:

4.1 The biochemical oxygen demand determination has its widest application in measuring waste loadings to treatment plants and in evaluating BOD-removal efficiency of such treatment systems.
4.2 This test is used to infer the general quality of the water and its degree of pollution by biodegradable organic matter. BOD is an indicator of the quality of a water body.
4.3 Urban runoff carries wastes from streets and sidewalks, nutrients from lawn fertilizers, leaves, grass, and paper from residential areas can all increase oxygen demand. Agricultural runoff can carry nutrients such as nitrogen and phosphates from fields, and runoff from animal feedlots carry fecal material into rivers.

5.0 Sample Collection, Preservation, and Holding Times:

5.1 The sample for BOD is collected with a minimum of headspace and refrigerated at $4^{o}C$ prior to analysis.
5.2 The maximum holding time prior to BOD incubation is 48 hours (CFR part 136). Since even a short time can significantly change the BOD results, the sample is set up for incubation as soon as practical after the sample has been taken.

6.0 Interferences:

6.1 Adjusting the temperature of the dilution water to 20°C prior to use is important for reproducible results.
6.2 Any chemical present in the sample that is detrimental to the seed bacteria would cause a negative interference (i.e. chlorine, acid, or alkaline).
6.3 See SOP I24 for interferences affecting the dissolved oxygen measurement.

7.0 Safety Issues: Some of the reagents used in the analysis are corrosive. Proper care must be taken to prevent contact. The use of gloves and safety glasses are essential when performing this test.

7.1 See SOP S01 – Concentrated Acids and Bases
SOP S03 – Spill Control Policy

8.0 Instrumentation/Equipment:

8.1 Incubator with temperature setting at 20 $\pm$ 1°C (excluding all light to prevent the possibility of photosynthetic production of dissolved oxygen).
8.2 300 mL capacity bottles with ground glass stoppers.
8.3 Plastic cup covers.
8.4 pH meter
8.5 Dissolved Oxygen Probe. (see I24)
8.6 General laboratory glassware: pipets, graduated cylinders.
8.7 Pall Life Sciences Supor 450 Membrane Filter 0.45 nm. 47 mm

Note: All glassware is cleaned immediately after use by thorough rinsing with three portions of D.I. water. If glassware still appears dirty, further steps are taken, such as 1:1 acid rinse. Glassware is always finished with a final D.I. rinse.

Periodically (as indicated by blank results) clean BOD bottles by filling with 1:1 HNO3, and rinsing thoroughly first with tap water and then finishing with three rinses of D.I. water. If a deeper clean is necessary heat on hotplate for 10 minutes and then cool prior to rinsing. *Always ensure that all acid is thoroughly removed from the bottle prior to use.*

9.0 Reagents and Standards:

Reagents are stored at room temperature for up to six months unless otherwise specified. Discard if there is any sign of biological growth. Standard Glucose-Glutamic Acid is made fresh daily.

9.1 Phosphate buffer solution: Dissolve 8.5 g KH_2PO_4, 21.75 g K_2HPO_4, 33.4 g $Na_2HPO_4 \cdot 7H_2O$ and 1.7 g NH_4Cl in 500 mL Nanopure water and dilute to 1 liter. pH should be 7.2.

9.2 Magnesium Sulfate solution: Dissolve 22.5 g $MgSO_4 \cdot 7H_2O$ in Nanopure water and dilute to 1 liter.

9.3 Calcium Chloride solution: Dissolve 27.5 g $CaCl_2$ or 36.5 g $CaCl_2 \cdot H_2O$ in Nanopure water and dilute to 1 liter.

9.4 Ferric Chloride solution: Dissolve 0.25 g $FeCl_3 \cdot 6H_2O$ in Nanopure water and dilute to 1 liter.

9.5 Standard Glucose (or Dextrose)-Glutamic Acid solution: Dissolve 0.15 g $C_6H_{12}O_6$ and 0.15 g $HO_2CCH_2 \cdot CH_2 \cdot CH(NH_2)CO_2H$ in Nanopure water and dilute to 500 mL. Place a stir bar into the flask and stir for 30 minutes. Prepare this standard fresh prior to each use. Plant 3 mL of solution into the BOD 300 mL bottle. The working standard solution is made at double strength, therefore 3 mL spiked into a BOD bottle represents a 2% dilution.

9.6 Sulfuric Acid Solution: Add 5 drops concentrated sulfuric acid to 100 mL of Nanopure Water.

9.7 0.25N NaOH Solution: Add 1 gram of NaOH to 100 mL of Nanopure Water.

9.8 Sodium Sulfite Titrant: Dissolve 0.7875 g of Na_2SO_3 into 500 mL of Nanopure water. Prepare fresh daily.

9.9 HACH Total Chlorine DPD crystals

9.10 Nitrification Inhibitor -contains 2-chloro-6-(trichloro methyl) pyridine (TCMP).

9.11 Reagent water: Nanopure

10.0 Procedure:

10.1 Sample Pretreatment

10.1.1 Test to determine if residual chlorine is present in the sample.

10.1.1.1 Place a few HACH Total Chlorine DPD crystals in a small container. Add approx. 3 mLs of sample and wait 3 minutes Pink color = chlorine present.

10.1.1.2 If present:

10.1.1.2.1 Place 200 mL of sample into a flask. Add 0.5 mL Sulfuric Acid, a few crystals of Potassium Iodide and a few drops of starch solution (see Dissolved Oxygen SOP I24). Titrate using Sodium sulfite solution until blue color disappears. Determine the amount of sodium sulfite solution needed to destroy the residual Chlorine in the amount of sample used for dilution.

10.1.1.2.2 Calculation -- Amount of Sodium sulfite solution from section 10.1.1.2 x Amount of sample needed for BOD divided by 100 = Amount of Sodium sulfite needed to destroy the

residual chlorine in the BOD sample. Add this amount to fresh sample.

10.1.2 Adjust pH to between 6.5 and 7.5.
10.1.2.1 Check the pH with pH paper or meter.
10.1.2.2 Adjust acidic samples with 0.25 N NaOH (1 g/100 mL DI) and basic samples with Sulfuric Acid (5 drops to 100 mL DI). Do not dilute by more than 1% (about 20 drops/100 mL). Use a higher concentration acid or base, if more than 1% is needed to neutralize the sample or adjust the pH of the diluted sample.

10.2 Sample Incubation and Analysis
10.2.1 Prepare BOD Dilution water: BOD water may be used up to 48 hrs after preparation.
10.2.1.1 Place 19 liters of Nanopure water into blue jug.
10.2.1.2 Add 1 mL each of Phosphate buffer, Magnesium Sulfate, Calcium Chloride, and Ferric Chloride solutions per liter of water.
10.2.1.3 Aerate water in refrigerator until temperature reads 19 - 21^{o}C. (Temperature of water will affect D.O. Regulate the temperature with a thermometer.)
10.2.1.4 Fill up a BOD bottle with the prepared dilution water for the unseeded blank.
10.2.1.5 Seed Nanopure water in the blue jug with 10 mL plant influent received in lab no longer than 24 to 48 hours ago. Preferably use plant influent that will be analyzed the same day and if possible, use influent as the duplicate source. When planting seed, pipet sample out of unshaken bottle being careful to pipet out the liquid portion near the top of the bottle. Mix seed into dilution water.
10.2.2 Arrange bottles so two-four dilutions of each sample can be made. If the determination of D.O. is by probe, then $D.O_1$ and $D.O_2$ are made from the same bottle, so only one bottle is needed for each dilution. Set up labeled bottles for blank, standards and Winkler standards.
10.2.3 Determine dilution amount and number of dilutions by using historical information, sample appearance, and odor. If uncertain, check with Manager. Clean samples require a 40% dilution.
10.2.4 Using wide tip pipette or graduated cylinder deliver desired amount of sample to each bottle by predetermined dilutions. Samples have been stored at 4^{o}C and may or *may not be 20^{o}C upon dilution.*
10.2.5 The dilution of the Lab Control is set to result in a concentration of 200 ppm. This is achieved by adding 3 mL of the Standard Glucose-Glutamic Acid solution into the 300 mL BOD bottle.
10.2.6 Fill each bottle with prepared water, preventing any air bubbles in the bottles. Form a water seal by allowing water to fill flare top to the lip of the bottle to ensure that no air will be trapped under stoppers after $D.O_1$ reading. Make sure that each batch (samples and QC) are poured from the same jug of dilution water.
10.2.7 Determine the initial dissolved Oxygen of samples. (see Dissolved Oxygen procedure SOP I24).

10.2.7.1 If the DO_1 is considerably lower (1 mg/L) than the other DO_1's and Blanks, then redo that sample using a lesser volume.

10.2.7.2 If the DO_1 is considerably higher (more than 9 mg/L), reduce DO to saturation by bringing another portion of sample to about 20 degrees Centigrade in a partially filled BOD bottle while agitating by vigorous shaking or by aerating.

10.2.8 Before placing ground glass stoppers into bottles, add more dilution water to top of bottle if necessary. Cover each bottle that is to be placed in the incubator with plastic caps. Put the covered bottles in the incubator.

10.2.9 After incubating samples for 5 days, determine the final Dissolved Oxygen (see Dissolved Oxygen procedure SOP I24).

10.3 Other BOD Products

10.3.1 BOD-dissolved - dissolved BOD: Filter sample though 0.45 micron filter prior to analysis. Do the same to a blank and duplicate sample. Filter 120 mL of Nanopure water and set up a 40% dilution for the method blank.

10.3.2 BOD-Carb – cabonaceous BOD: Add 0.16 g Nitrification Inhibitor (containing 2-chloro-6-(trichloro methyl) pyridine -TCMP) to BOD bottle containing sample. Do the same to a blank, LCS, and duplicate sample.

10.4 Other matrices such as solids and sludges: Discuss with Manager.

10.4.1 Perform a duplicate on a solid or sludge sample.

11.0 Quality Control:

Note: See also ESB SOP Q01 for general QC requirements

11.1 Method Blanks are analyzed with every 20 samples or once per batch, whichever is more to check for possible interference from seed and reagents. Method Blanks are reported per matrix type. The target range for unadjusted blank depletion $(B_1 - B_2)$ is 0.6 - 1 mg/L DO. Analysts should double check DO readings and notify a Manager if blank readings exceed the target range. See section 12.2.3.

11.2 Duplicate analyses are performed at a frequency of one in 20 samples, per matrix type or once per batch, whichever is more.

11.2.1 The relative percent difference is calculated for the duplicate samples run as follows:

$$RPD = \frac{ABS(Run\ 1 - Run\ 2)}{(Run\ 1 + Run\ 2) / 2} \times 100$$

11.2.2 Duplicates must be within a maximum RPD of 20%.

11.3 A laboratory control standard is analyzed every 20 samples or once per batch, whichever is more and reported for every matrix type.

11.3.1 The method target range for historical limits is 85-115% based on a series of laboratory studies which gave the following results: "For the 300 mg/L mixed primary standard, the average 5-d BOD was 198 mg/L with a standard deviation of 30.5 mg/L). Lab control standards outside the acceptance range cause the analysis to be put into question. Troubleshooting must be performed to attempt to identify the problem. The Winkler LCS may be used to accept a sample batch. The Manager, QA Manager or Laboratory Technical Director is consulted to determine if data is acceptable and reportable. If data is reported in a batch without an acceptable LCS, all client reports must be flagged with NLbod. See ESB SOP Q06 for more details.

11.4 Probe Blank: An unseeded blank is analyzed with each batch to monitor probe performance. The target range for the blank is <0.4 mg/L. An elevated result may be an indication of poor quality dilution water. Please consult your Manager.

11.5 An MDL study is completed whenever major equipment or procedural changes are made. A standard Glucose-Glutamic Acid solution is prepared as described in section 9.5. This is calculated as a 100% solution. The dissolved oxygen depletion of seven replicates is tabulated. See QA Manual for calculation. Results must be below the reporting limit.

11.5.1 LOD Verification: On a yearly basis, an aliquot of water is spiked at a concentration level of 4 mg/L. This is calculated as a 100% solution. The sample is analyzed. Analyte response must be greater than method blank response to verify the Level of Detection or MDL.

11.6 Initial Demonstration of Capability: Prior to analysis of samples or when a significant change is made to the method, an Initial Demonstration of Capability Study is performed. This is accomplished by analysis of four replicates of a Lab Control Sample for BOD and cBOD. Plant 3 mL of Standard Glucose-Glutamic Acid solution from 9.5 into a BOD bottle. This is calculated as a 2% solution. Acceptance ranges are 80-120% with a maximum %RSD of 20.

11.7 Demonstration of Continuing Proficiency: On an annual basis, each analyst must turn in valid LCS data from four consecutive batches or results from a successful Performance Evaluation Study. LCS percent recovery must meet laboratory prescribed acceptance criteria. Relative standard deviation between the four replicates must be less than or equal to 20%. Performance Evaluation Studies are performed twice a year.

12.0 Calculations:

12.1 For seeded samples:

$$\text{BOD mg/L} = \frac{(D.O._1 - D.O._2) - (B_1 - B_2)\, f}{W} \times 100$$

or

$$\text{BOD mg/L} = \frac{\text{Sample depletion} - \left[\text{blank depletion} \times \frac{\text{mL dilution water in sample}}{300\ \text{mL}}\right]}{\text{percent sample}} \times 100$$

Where:	Example
$D.O_1$. = D.O. of diluted sample immediately after preparation.	8.5
$D.O_2$. = D.O. of diluted sample after a 5 day incubation at 20°C.	4.5
B_1. = D.O. of seeded blank immediately after preparation.	8.7
B_2. = D.O. of seeded blank after a 5 day incubation at 20°C.	8.1
W = % of sample in BOD bottle.	40
f = Ratio of seed in diluted sample to seed in seed control	180/300

Example:

$$\text{BOD mg/L} = \frac{(8.5-4.5) - [(8.7-8.1)\,(180/300)\}}{40} \times 100 = \frac{4.0 - 0.36}{40} \times 100 = 9.1\ \text{mg/L}$$

12.2 Blank Depletion: $(B_1 - B_2)$

12.2.1 For the above equation, use the blank depletion of the blank bottle poured from the same jug of dilution water and therefore associated with that batch of samples. (If there are two blanks associated with a batch, use the blank with the smallest depletion).

12.2.2 Enter the Method Blank into LIMS by taking the batch blank depletion $(B_1 - B_2)$ adjusted by the historical blank depletion average (0.6 for BOD, 0.4 for CBOD and DBOD) and entered as 100%.

12.2.3 The Method Blank is considered to have failed if the unadjusted blank depletion $(B_1 - B_2)$ is greater than 1 mg/L. If the depletion is greater than 1 mg/L:

12.2.3.1 Adjust sample results using a $(B_1 - B_2)$ of 1 mg/L.

12.2.3.2 Enter the method blank result calculated from section 12.2.2 with a qualifier QBLK

12.2.3.3 Use the following equation to determine the contamination level in the sample. Calculate the blank contamination for each sample dilution used in the final BOD result. Average the contamination values. Qualify samples (N-Bblk) with reportable results that are less than 10 times the average blank contamination level.

$$(B_1 - B_2) - 1.0 \times \frac{\text{Volume dilution water in sample}}{\text{Volume of sample}} = \text{Blank Contamination}$$

13.0 Reporting:

13.1 The following criteria is followed when choosing a reportable result:

13.1.1 Residual DO of at least 1 mg/L.
13.1.2 DO depletion of at least 2 mg/L. (prior to blank correction)
13.1.3 No evidence of toxicity (depletion increasing as sample volume decreases) or obvious anomaly. Refer to Historical data, COD, TOC, or TSS result if needed.

13.2 All reportable results are averaged
13.2.1 Report the average result.
13.2.2 Calculate the reporting limit based on the ratio of dilution water in the sample and volume of sample used:

$$2 \times \frac{300 \text{ mL}}{\text{*Volume of sample (mL)}} = \text{Adjusted RL}$$

*Values are taken from the sample dilution, included in the average calculation that contains the highest volume of sample.

13.3 If none of the dilutions meet the criteria in section 13.1, the manager is consulted to determine the most appropriate course of action.

13.3.1 If residual DO is < 1 mg/L then too much sample was used. Qualify the sample as follows:
13.3.1.1 A “greater than” result will be reported using the _BOD qualifier. (Use 2 significant figures.)
13.3.1.2 The greater than value is calculated as the highest value that the sample could have been had its smallest dilution depleted to 1.0 mg/L.

13.3.2 If DO depletion < 2 mg/L. then insufficient sample was used. Report the result of the sample dilution that contains the highest volume of sample. Do not average. Qualify the sample as follows:
13.3.2.1 Report data using the _BOD qualifier. This qualifier allows the result to be displayed even though it is less than the elevated reporting limit. (Use 2 significant figures.) Add N-BOD to explain that results did not meet method calculation criteria and the reported result is an estimated value only.
13.3.2.2 Exceptions to section 13.3.2.1 include
13.3.2.2.1 Samples with a dilution of 10% or greater; in those cases, no qualifications are necessary and the client will see ND at an RL of 20 mg/L or less.

13.3.2.2.2 Samples that meet calculation criteria but the adjusted depletion < 2, making the final result less than the calculated RL; in those cases, enter the BOD result using the _BODJ qualifier. Enter the result with two significant figures followed by a "J" (ex 96J). Add the qualifier N-BODJ. The result will be reported to two significant figures and followed by a "J" on the final report.

13.3.2.3 If no BOD result is available, (i.e sample was not planted or probed), add the qualifier N-Nae, result not available due to lab error.

13.3.3 Each of the above scenarios (except section 13.3.2.2) requires that a BOD cancellation form be filled out, with estimated results and client information.

13.3.3.1 Print a copy of the bench sheet
Make a copy of every cancellation form for peer review. Give the original to the project manager associated with that client.

13.4 Turn in for peer review:

13.4.1 Original, completed data page
13.4.2 Copy of bench sheet
13.4.3 Copies of BOD cancellation forms (if applicable)
13.4.4 Completed BOD peer review sheet

13.5 The reporting units for this analysis are mg/L.

13.6 LIMS entry

13.6.1 Enter DO readings and other analysis info into spreadsheet.

13.6.2 Review for accuracy.

13.6.3 Export results and enter into Element.

13.6.4 Analyzed date and time is documented as the date and time the last probe reading in the run was taken.

13.6.5 The analyst is defined as the person who planted the sample.

13.7 *Customers can refer to this SOP Section 5 for temperature and duration of storage information.*

14.0 Definitions: See SOP Q15 – SOP Definitions
Seed Control = Method Blank

15.0 Corrective Action For Out of Control Or Unacceptable Data:
See SOP Q06 – Corrective Action

16.0 Pollution Prevention and Waste Management:

16.1 Waste is discharged directly to sewer.
16.2See SOP S05 – Neutralization Procedure for Acid and Alkaline Wastes
SOP S07 – Pollution Prevention

17.0 References:
Standard Methods for the Examination of Water and Wastewater, APHA, AWWA, WEF, 22nd Edition. 5210B
EPA 405.1, EPA Methods for the Chemical Analysis of Water and Wastes.

Note: All *italicized items* are an indication of a variation from the method.

Approved by: ___Julia Sudds_____ Date: _____04/03/14____ ___

Chemical Oxygen Demand
Standard Operating Procedure
Babcock Laboratories, Inc.
SM 5220D
Effective Date: ___04/07/14_____

1.0 Scope and Application

1.1. This method covers the determination of COD in surface waters, domestic and industrial wastes.

1.2. This method may also be used for solid samples if both client and regulator agree to this variation.

1.3. The applicable range of the colorimetric method is 10-500 mg/L. The reporting limit is 10 mg/L.

1.4. Environmental Relevance:

1.4.1. Oxidizable chemicals (such as reducing chemicals) introduced into a natural water will similarly initiate chemical reactions. The chemical reactions create what is measured in the lab as chemical oxygen demand (COD).

1.4.2. COD tests measure the relative oxygen depletion effect of a waste contaminant. This test determines whether or not a specific wastewater will have a significant adverse effect upon fish or upon aquatic plant life.

2.0 Summary of Method

2.1 Sample, blanks and standards in sealed tubes are heated in an oven block digester in the presence of dichromate at 150^{o}C. After two hours, the tubes are removed from the oven or digester, cooled and measured spectrophotometrically at 620 nm.

3.0 Sample Handling and Preservation

3.1 Collect the samples in glass bottles if possible. Use of plastic containers is permissible if it is known that no organic contaminants are present in the containers.

3.2 Samples should be analyzed as soon as possible or preserved with sulfuric acid to a pH < 2 and maintained at 4^{o}C until analysis. Analysis performed on unpreserved samples are qualified N_UNP. Analysis must take place within

28 days. Samples are kept in plastic bottles or snap caps in the refrigerator on the sulfuric preserved cart.

3.3 Upon receipt to the laboratory, samples containing suspended material are homogenized using a tissue tearer.

4.0 Interferences

4.1 Chlorides—Chlorides are quantitatively oxidized by dichromate and represent a positive interference. Mercuric sulfate is added to the digestion tubes in the Hach reagent to complex the chlorides. This is effective for chloride levels of 2000 mg/L or less. High concentrations of chloride will precipitate and become turbid when added to the COD solution. Samples with higher amounts of chloride are *diluted* until the chloride level is no longer an interferent.

4.2 Discard vials that are scratched or blemished.

5.0 Safety

5.1 Safety goggles and gloves are recommended during this procedure since the reagent vials contain sulfuric acid which will heat up when the sample is added as well as other hazardous chemicals such as dichromate and mercury. These chemicals share the potential to be highly toxic or hazardous. Consult the MSDS that is located in the file cabinet in the study.

5.2 See SOP S01 – Concentrated Acids and Bases
SOP S03 – Spill Control Policy

6.0 Apparatus

6.1 Biospec Tissue Tearer

6.2 Drying oven or block digestor, 150^{o}C. Inst. ID# 70.

6.3 HACH Digestion vials containing prepared digestion solution for COD with a range of 0-1500 ppm.

6.4 Spectrophotometer – Genesys 10uv. A Inst. ID# 227

6.5 Standard laboratory glassware: volumetric flasks, beakers, graduated cylinders, pipettes.

Note: All glassware is cleaned immediately prior to and after use by thorough rinsing with three portions of D.I. water. If glassware still appears dirty, further steps are taken, by use of one of the following: Alconox and hot water, 1:1 acid rinse, acetone or appropriate solvent rinse. Glassware is always finished with a final D.I. rinse.

7.0 Reagents and Standards

7.1 Stock potassium acid phthalate (potassium biphthalate or potassium Hydrogen Phthalate) The salt is stored at room temperature for up to 10 years from date received.

7.1.1 LCS/MS Intermediate Standard (500 ppm) -Dissolve 0.425 g in Nanopure water and dilute to 1 liter. 1 mL = 250 mg/L COD, 0.2 mL = 50 mg/L COD. Store LCS solution in the refrigerator in an amber bottle for up to 3 months.

7.1.1.1 Midlevel LCS 250 ppm: 1 mL of above intermediate and 1 mL Nanopure into COD vial, made fresh each run

7.1.1.2 High Level LCS 500 ppm: 2 mLs of above intermediate into COD vial, made fresh each run. (High CCV)

7.1.1.3 Working MS 125 ppm: 0.5 mL of above intermediate, 0.5 mL of Nanopure, and 1 mL sample into COD vial, made fresh each run.

7.1.2 Calibration Intermediate Standard (500 ppm): Follow above procedure using a second source of potassium acid phthalate. Store STD Intermediate in the refrigerator in amber bottles for up to 3 months.

7.1.2.1 Dilute Intermediate Standard further to create a seven point curve. Previous curves have been prepared at the following levels:

Conc.	Nano	Cal Std.
0.0 mg/L	2 mL	0 mL
10 mg/L	1.96 mL	0.04 mL
62.5 mg/L	1.75 mL	0.25 mL
125 mg/L	1.5 mL	0.5 mL
250 mg/L	1.0 mL	1.0 mL
375 mg/L	0.5 mL	1.5 mL
500 mg/L	0 mL	2mL

7.1.2.2 Single point calibrator 250 ppm: 1 mL of 500 ppm Calibration Intermediate standard (made fresh when recalibration is necessary) and 1 mL Nanopure into COD vial.

7.2 Digestion Solution: Purchased pre-made reagent from HACH. Vials are stored at room temperature until manufacturer's specified holding time.

7.3 Reagent water: Nanopure acidified to a pH of <2 with sulfuric acid.

8.0 Colorimetric Procedure

8.1 Uncap and number 25 HACH vials (MB), 2, 3, 4, 5, etc.

8.2 Turn on COD reactor to infinity to warm up.

8.3 Add 2 mLs of reagent water to MB vial (if prepping, see 8.5), 1 mL of the LCS intermediate standard to LCS vial(#2), 0.5 mL of the LCS intermediate standard to the MS and MSD vials, and 2 mLs of the LCS intermediate standard to the high level LCS vial.

8.4 Add 2 mLs of aqueous sample to corresponding numbered vial. Add 1.5 mL of sample chosen for spike (if 2 mLs used) to the MS and MSD vials. A 1 mL autopipetter with the end of the tip clipped off is used for QC samples and samples where 1 mL or less is used. Use a 5 mL autopipetter with the end of the tip clipped off for all other samples. A serial dilution should be used for measurements less the 500 uL.

8.4.1 For solid samples weigh out approximately 0.25 g of solid sample to the corresponding numbered vial, as well as the MS and MSD vials. Add the appropriate amount of reagent water to bring up to a final volume of 2 mLs.

8.5 If a sample is dirty or turns a greenish color when added to the vial less sample must be used. If sample turns turbid or precipitates see section 4.1. Choose dilutions according to sample color, clearness, and odor. Mark the amount of sample used on the COD prep sheet. Add reagent water to bring up QC samples and any diluted samples up to 2 mLs. Verify that the liquid level is the same throughout the batch.

8.6 Cap tightly and shake to mix layers. A thorough mixing is necessary before heating to prevent local heating of sulfuric acid layer on bottom of tube that may cause an explosive reaction.

8.7 Process standards and blanks exactly as the samples.

8.8 Place in oven or block digester at 150°C for two hours. Set timer for 120 minutes. Leave tubes in reactor for approximately 20 minutes after timer goes off or use tool to remove them from the digester and place in rack to cool.

8.9 Once in rack, invert cooled samples several times. Make sure vials are cool to the touch before measuring absorbance. Dislodge solids that adhere to

container by gently tapping. If sample is turbid, let it settle longer, use a centrifuge, or reanalyze with less sample volume.

8.10 Measure color intensity by setting the spec to read at *620 nm*. On the spec select "Set nm" and enter 620. This wavelength was determined to be optimal for our laboratory conditions.

8.11 A method blank that is digested and read must be analyzed once a week and for each new lot of Hach tubes. This digested reagent blank is checked with each batch per sec. 8.12 and used to zero the spectrophotometer. *This may be kept as long as it is stable*. Store it in a dark place.

8.12 Check the digested reagent blank by placing a vial containing a minimum of 5 mLs of Nanopure into the spec. Set transmittance to 100% by selecting "Change Mode" until %T appears and then select "Measure Blank." Then read the transmittance of the reagent blank. Record this value on the data page. If the transmittance has changed more than 2% from the original reading or a new lot of vials is started, a new reagent blank must be digested and used to zero the spec. (per manufacturers instructions – Hach COD Reaction Model 45600 pg51).

8.13 To zero the spec using the digested reagent blank: Select "Change Mode" until absorbance appears. Select "Measure Blank" (which zeros the spec).

8.14 Wipe the first vial off with a damp tissue and dry off making sure vial is clean and free of fingerprints or scratches. Place vial in the spec with the label facing forward. Read the absorbance and remove the vial. Repeat with remaining vials.

9.0 Product Variation

9.1 Dissolved COD results are obtained by filtering the sample thru a 0.45 micron filter and then proceeding with the analysis in step 8.4. Refer to A06 for more information.

NOTE: If sample(s) are filtered, filter all QC samples: MB, LCS, MS/MSD, and the high CCV sample.

10.0 Calibration and Calculation

10.1 Five calibration standards are required by the method. In general calibration standards are prepared at seven concentration levels including zero. A new curve is prepared on a *yearly* basis covering the referenced concentration range (see 7.1.2.1).

10.2 An acceptable curve yields an $r^2 = 0.99$ or better.

10.3 A regression equation is generated from the curve.

10.4 Results are calculated using the regression equation. See most current curve for proper equation.

10.5 Samples that read above the high standard must be redone at a higher dilution.

10.6 If result appears inconsistent with client i.d., re-read absorbance by turning vial a half turn.

10.7 Immediately after calibration a 250 mg/L lab control serves as the ICV. It must be within 95% - 105% of the expected value or the calibration curve is redone.

10.8 See below for CCV information.

11.0 Quality Control

Note: See also ESB SOP Q01 for general QC requirements

11.1 The 250 mg/L lab control is used as an ICV and CCV. It is analyzed every twenty samples per matrix type or one per analysis batch whichever is greater. If recovery is not within 95% - 105% of the expected value either the analysis is repeated or it is recalculated based on a single point calibrator.

11.2 A MS/MSD is analyzed every twenty samples per matrix type or one per analysis batch whichever is greater. The acceptance range is 80% - 120% of the expected value and 75%-120% for solids/sludges. The relative percent difference between the spikes must be no greater than 20%. MSD percent recovery must be acceptable to satisfy MS frequency requirements.

11.3 The 500 mg/L lab control is used as the ending CCV. It is analyzed at the end of the batch. If recovery is not within 95% - 105% of the expected value a new one is digested and reanalyzed. If the redigested CCV fails, a 250 mg/L calibrator is digested from a non LCS source. It is used as a single point calibrator to recalculate the rest of the batch as follows:

$$\text{Samp. Conc.} = \frac{(\text{Samp. Abs.})(\text{Conc. Single Pt. Cal.})}{\text{Single Pt. Cal. Abs}}$$

11.4 The method blank is run once a week. It must be below the reporting limit. Sample results at or above the reporting limit must be accompanied with a qualifier if the method blank exceeds a concentration greater than 1/10 the measured raw concentration of the sample. Blank results below the reporting limit are considered to be ND and will not require a note.

11.5 An MDL study is completed whenever major equipment or procedural changes are made. Standards are spiked at the reporting limit or 2.5-5 times the estimated MDL and a minimum of seven replicates is analyzed. See QA Manual for calculation. Results must be below the reporting limit.

11.5.1 LOD Verification: On an annual basis, an aliquot of water is spiked at a level of 2-3 times analyte MDL. The sample is analyzed. Analyte response must be greater than method blank response to verify the Level of Detection or MDL.

11.6 Initial Demonstration of Capability: Prior to analysis of samples or when a significant change is made to the method, an Initial Demonstration of Capability Study is performed. This is accomplished by analysis of four replicates of a QC sample made at a concentration 10 times the MDL to midrange of the curve. The average percent recovery of the QC samples must be 80-120% with a maximum %RSD of 20.

11.7 Demonstration of Continuing Proficiency: On an annual basis, analysts must turn in valid LCS data from four consecutive LCS samples or results from a successful Performance Evaluation Study. LCS percent recovery must meet laboratory prescribed acceptance criteria. Relative standard deviation between the four replicates must be less than or equal to 20%. Performance Evaluation Studies performed twice a year.

12.0 Definitions: See SOP Q15 – SOP Definitions

13.0 Corrective Action For Out of Control or Unacceptable Data:

13.1 See SOP Q06 – Corrective Action

14.0 Reporting:

14.1 Results are reported as mg/L.

14.2 The reporting limit is 10 mg/L. Refer to Element for MDL information.

14.3 Prepared time is documented as the time the samples are placed in the block digester.

14.4 Analysis time is documented as the time the absorbance of the last sample in the batch was read.

14.5 The results are reported to 2 significant figures.

15.0 Pollution Prevention and Waste Management:

14.1 See SOP S05 – Neutralization Procedure for Acid and Alkaline Wastes
SOP S07 – Pollution Prevention

14.2 Waste Disposal: Each sample vial's contents must be poured into a COD only waste receptacle and the empty vials must be discarded only into the GLASS ONLY waste receptacle. The COD only waste receptacle is handled by a Waste Disposal company.

16.0 Method Performance

Refer to MDL studies, Initial Demonstration of Capability Studies, and laboratory control charts maintained in the QA Office.

References

Standard Methods For the Examination of Water and Wastewater APHA, AWWA, WEF 22nd Edition 5220 D.
Instrument Manual for COD Reactor Model 45600

Note: All *italicized items* are an indication of a variation from the method.

Approved by: _____Julia Sudds_____ Date: __ 4/3/14_____ __

Standard Operating Procedure
Babcock Laboratories, Inc.
EPA 300.0/EPA ML 300.0
Effective Date 4/4/2014

TITLE: The Determination of Inorganic Anions in Water by Ion Chromatography

ANALYTE:	CAS #	MCL (mg/L)
Chloride Cl	7782-50-5	250/500/600*
Nitrate NO_3	14797-55-8	45
Sulfate SO_4	14808-79-8	250/500/600*

Non NELAP Analytes:
Para-Chlorobenzene Sulfonic Acid PCBSA 98-66-8
*Secondary MCL (non-health limit) recommended/upper/short term

1.0 Scope and Application

1.1. This method covers the determination of the following inorganic anions.

		RL, mg/L
1.1.1.	Chloride	1
1.1.2.	Nitrate-N / Nitrate	0.2 / 1
1.1.3.	Sulfate	0.5
1.1.4.	PCBSA	10

1.2. The matrices applicable to each method are shown below:
Drinking water, surface water, mixed domestic and industrial wastewaters, groundwater, reagent waters, solids (after extraction 11.11).

1.3. The working range for these analytes is as follows:

1.3.1.	Chloride	1-250 mg/L
1.3.2.	Nitrate/NO_3-N	1-100 mg/L/0.2257-22.57mg/L
1.3.3.	Sulfate	0.5-250 mg/L
1.3.4.	Perchlorate	0.004-0.25 mg/L
1.3.5.	PCBSA	1-100 mg/L

1.4. This method is recommended for use only by or under the supervision of analysts experienced in the use of ion chromatography and in the interpretation of the resulting ion chromatogram. Each analyst must demonstrate the ability to generate acceptable results with this method, using the procedure described in Section 10.2.

1.5. When this method is used to analyze unfamiliar samples for any of the above anions, anion identification should be supported by the use of fortified sample

matrix covering the anions of interest if the identity of an analyte is in question.

1.6. Environmental Relevance:

1.6.1. Nitrate is regulated as a health concern because it has been identified as a cause of methemoglobinemia. In freshwater or estuarine systems nitrate can reach high levels that can potentially cause the death of fish. Over 30 ppm of nitrate can inhibit growth and impair the immune system of fish.

1.6.2. Chloride increases the electrical conductivity of water and increases its corrosivity. In metal pipes, chloride reacts with metal ions to form soluble salts thus increasing levels of metals in drinking water.

1.6.3. Sulfate may have a laxative effect that can lead to dehydration. With time, the symptoms will disappear as people adapt to the sulfate in the water. Sulfate can also cause scale buildup in water pipes and corrode plumbing.

2.0 Summary of Method

2.1 A *25 µL* aliquot of sample is injected into an eluent stream and passed through a series of ion exchangers. The system is comprised of a guard column, separator column, and suppressor device. These separate the ions based on their affinity for a low capacity, strongly basic ion exchanger. They are then directed onto a strongly acidic cation exchanger where they are converted to their highly conductive acidic forms. The conductivity of these acid forms is measured. Identification is based on retention time. Quantitation is based on peak height or peak area.

2.2 In order to use this method for solids an extraction procedure must be performed (See Sec 11.11).

3.0 Definitions (see SOP Q15 for definitions)

4.0 Interferences

4.1 Interferences can be caused by substances with retention times that are similar to and overlap those of the anion of interest. Large amounts of an anion can interfere with the peak resolution of an adjacent anion. Sample dilution and/or spiking can be used to solve most interference problems.

4.2 Method interferences may be caused by contaminants in the reagent water, reagents, glassware, and other sample processing apparatus that lead to discrete artifacts or elevated baseline in ion chromatograms.

4.3 Samples that contain particles larger than 0.45 microns and reagent solutions that contain particles larger than 0.20 microns require filtration to prevent damage to instrument columns and flow systems. Caution: filtration may remove perchlorate.

4.4 The acetate anion elutes early during the chromatographic run. The retention times of the anions also seem to differ when large amounts of acetate are present. Therefore, this method is not recommended for leachates of solid samples when acetic acid is used for pH adjustment.

5.0 Safety

5.1 Normal, accepted laboratory safety practices should be followed during reagent preparation and instrument operation. No known carcinogenic materials are used in this method. Each chemical should be regarded as a potential health hazard and exposure should be as low as reasonably achievable.

5.2 Each laboratory is responsible for maintaining a current awareness file of OSHA regulations regarding the safe handling of the chemicals specified in this method. A reference file of Material Safety Data Sheets (MSDS) is available to all employees in the filing cabinet in the hallway across from the QA office.

5.3 See SOP S01 – Concentrated Acids and Bases
SOP S02 – Compressed Gas Cylinder Handling
SOP S03 – Spill Control Policy

6.0 Apparatus and Materials

6.1 Balance - Analytical, capable of accurately weighing to the nearest 0.0001 g.

6.2 Ion chromatograph - Analytical system complete with ion chromatograph and all required accessories including syringes, analytical columns, compressed gasses and detectors.

6.2.1 IC: Dionex DX –120 1, DX ICS-1, Thermo Scientific/Dionex ICS 1100-2 and Dionex ICS 2100 (utilized only as backup)
Pump Rate: 2.0 mL/min.

Eluent: as specified in 7.3
Sample Loop: 25 uL

6.2.2 Data Handling: Pentium Processor with Chromeleon (version 6.8 & 7.0) software on Windows NT platform.

6.2.3 Printer: HP Laser Jet 4200, 4300

6.2.4 Autosampler: Alcott Micromeritics 728, 709 and Thermo/Dionex AS-DV

6.2.5 Anion guard column: AG22 4mm Dionex Guard Column (or equivalent). A protector of the separator column. If omitted from the system the retention times will be shorter. Usually packed with a substrate which is the same in the separator column.

6.2.6 Anion separator column: AS-22 4mm Dionex Column (or equivalent)

6.2.7 Anion suppressor column: Anion self-regenerating ASRS-11.

6.2.8 Detector – Conductivity cell

6.3 Standard laboratory glassware: volumetric flasks, beakers, graduated cylinders, pipets.

Note: All glassware is cleaned immediately prior to and after use by thorough rinsing with three portions of D.I. water.

7.0 Reagents and Consumable Materials

7.1 Sample bottles: Glass or polyethylene, sufficient volume to allow replicate analyses of anions of interest.

7.2 Reagent water: Nanopure, free of the anions of interest. Water should contain particles no larger than 0.20 microns with a conductance of <0.1 uS/cm.

7.3 Eluent solution: Dissolve 11.76 g sodium bicarbonate ($NaHCO^3$) and 47.7 g of sodium carbonate (Na^2CO^3) in nanopure water (7.2) and dilute to 1 liter. Store at room temperature for up to 6 months.

7.4 Stock standard solutions 1000 mg/L and 10,000 mg/L solutions: Stock standard solutions are purchased as certified solutions. Store standards at 4^oC until manufacturer specified holding date. Once opened, the expiration date is 1 year from date opened as long as this does not exceed manufacturer's original expiration date.

7.5 Working standard solutions: See section 9.5 for concentrations and recipes. Working standards are stored at room temperature. Calibration standards are stored for up to *3 months.* LCS solutions are prepared weekly.

8.0 Sample Collection, Preservation and Storage

8.1 Samples are collected in scrupulously clean glass or polyethylene bottles.

8.2 Sample preservation and holding times for the anions that can be determined by this method are as follows:

Analyte	Preservation	Holding Time
Chloride	None required	28 days
Nitrate (Unchlorinated)	Cool to 4^{o}C	48 hours
Nitrate-N/Nitrite-N		
chlorinated	Cool to 4°C	14 days
combined	conc. H_2SO_4 pH<2*	28 days
Sulfate	Cool to 4°C	28 days
PCBSA	Cool to 4°C	28 days

8.3 The method of preservation and the holding time for samples analyzed by this method are determined by the anions of interest. In a given sample, the anion that requires the most preservation treatment and the shortest holding time will determine the preservation treatment. It is recommended that all samples be cooled to 4°C and held no longer than 28 days. *If a preserved sample is used it must be neutralized prior to injection.

8.4 If a preserved sample is going to be analyzed to report a combined Nitrate-N/Nitrite-N result and extend the holding time to 28 days* the following steps must be taken:

- Since the 2 analytes are reported together, one must look for the nitrite-N peak on the chromatogram:

1. Inject a 0.1ppm nitrite-N standard with the sample run.
   i) Spike 10 uL of 1000 ppm Nitrite-N stock standard into 100 mL of cal std#1.
2. Examine the sample chromatogram (by zooming in) to determine if Nitrite-N is detected in the sample at or above the 0.1 ppm reporting limit.
   i) If detected $\geq$ 0.1 ppm:
      (a) Calculate the concentration for the Nitrite-N peak in the sample using the 0.1 ppm standard. (If Nitrite peak is elevated a 0.5 ppm standard may be used.)
      (b) 0.1 ppm x (height of sample peak/height of 0.1 ppm peak) = Nitrite-N as ppm in sample
   ii) Add above concentration to the Nitrate-N answer. In this case the Total Nitrogen will need to be adjusted by removing the colorimetric Nitrite-N result, so that Nitrite-N is not added into the Total Nitrogen result twice.

iii) If the observed response is <0.1 ppm then report the Nitrate-N answer as is and allow the Total Nitrogen to calculate as normal.

- In all of these cases, the colorimetric nitrite result will still be reported as normal
- Attach the qualifier *N_HTsa to the nitrate result.

* N_HTsa note: "Determined on Sulfuric Acid preserved sample - holding time extended to 28 days per EPA Method 300.0. Result is expressed as Nitrate/Nitrite- Nitrogen."

9.0 Calibration and Standardization

9.1 Calibrators at five levels and a blank are required for each analyte of interest. Generally calibration standards are prepared at eight concentration levels, plus a blank, by adding accurately measured volumes of one or more stock standards to a volumetric flask and diluting to volume with reagent water as specified in section 9.5.5. The blank (zero point) is included but not forced through zero. (See sections 9.5.5 and 9.5.6). A quadratic curve fit is used.

9.2 An acceptable curve has an $r^2 \geq 0.99$.

9.3 Using *25uL* injections of each calibration standard tabulate peak height or area responses against the concentration. The results are used to prepare a calibration curve for each analyte. During this procedure, retention times must be recorded.

9.4 The calibration curve is verified on each working day, prior to sample analysis after every 10 samples and at the end of the run. *Under circumstances where the analytical run continues during the absence of analyst supervison, such as during nights and weekends, two CCVs may be loaded in succession every 10 samples to prevent QC criteria failure for samples bracketed by acceptable CCVs and analyzed during the run where a CCV may not meet criteria.* The IPC, an Instrument Performance Check, is essentially equivalent to the ICV/CCV required in other methods. If the response for any analyte varies from the expected values by more than 10%, the test must be repeated, using fresh calibration standards. If the results are still out of range, a new calibration curve must be prepared for that analyte and results that are not bracketed by working QC must be qualified or reanalyzed. *If the retention time for any analyte varies outside the curtain window, reset the retention time to the current CCV value and reprocess the run.*

9.5 Standard Concentrations and Recipes:

9.5.1 ICV/IPC Standard: (Initial Calibration Verification/Instrument Performance Check) Solution is prepared weekly.

Analyte	**Conc.**	**Acceptance Range %**
Cl	50 ppm	90-110 (45 ppm-55 ppm)
NO_3	50 ppm	90-110 (45 ppm-55 ppm)
SO_4	50 ppm	90-110 (45 ppm-55 ppm)

9.5.2 CCV/IPC Standards: For recipes see Calibration Standard #5 (Mid) and #8 (High). Standards have a holding time of 3 months.

Analyte	**Conc.**		**Acceptance Range %**
	Mid	High	
Cl	50	250 ppm	90-110
NO_3	50	100 ppm	90-110
SO_4	50	250 ppm	90-110

9.5.3 PCBSA CCV standards: For recipes see Calibration Standard #4 and #5. Standards have a holding time of 1 year.

Analyte	**Conc.**		**Acceptance Range %**
	Mid	High	
PCBSA	50	100 ppm	90-110

9.5.4 Low-level Check Standards: Holding time of 3 months

9.5.4.1 Low Level CCV (Standard #3): A low level CCV, Chloride, Nitrate, and Sulfate all at 20 ppm is analyzed at the end of each analytical run. If the Low-Level CCV varies by more than 15%, samples having results less than 35ppm are reanalyzed.

9.5.4.2 RL Check (Standard #1): A standard at the reporting limit is analyzed at the end. This is used to validate

samples reported as ND and to evaluate instrument sensitivity should calibration checks show a low bias. The standard should have a signal greater than the method blank.

9.5.5 Calibration Standards (Cl, NO_3, SO_4): The eight standards below are made from a 1000 ppm stock solution into a volumetric flask or a graduated cylinder and brought up to volume with Nanopure water. Holding time *3 months*.

Std #1		mL into 1L
Cl	1 ppm	1.0
NO_3	1 ppm	1.0
SO_4	0.5 ppm	0.5

Std #2		mL into 50 mL
Cl	10 ppm	0.5
NO_3	10 ppm	0.5
SO_4	10 ppm	0.5

Std #3		mL into 50 mL
Cl	20 ppm	1.0
NO_3	20 ppm	1.0
SO_4	20 ppm	1.0

Std #4		mL into 50 mL
Cl	35 ppm	1.75
NO_3	35 ppm	1.75
SO_4	35 ppm	1.75

Std #5		mL into 200 mL
Cl	50 ppm	10
NO_3	50 ppm	10
SO_4	50 ppm	10

Std #6		mL into 50 mL
Cl	75 ppm	3.75
NO_3	60 ppm	3.0
SO_4	100 ppm	5.0

Std #7		mL into 50 mL
Cl	100 ppm	5.0
NO_3	75 ppm	3.75
SO_4	150 ppm	7.5

Std #8		mL into 200 mL
Cl	250 ppm	50
NO_3	100 ppm	20
SO_4	250 ppm	50

9.5.6 PCBSA Calibration Standards

9.5.6.1 Stock Standard Salt: 4-Chlorobenzenesulfonic acid purchased from a certified vendor. Store at room temperature for up to 10 years from date received.

9.5.6.2 Intermediate Standard 100ppm: Weigh 0.1g of 4-Chlorobenzenesulfonic acid into 1L of Nanopure. Store this standard at room temperature for up to one year.

9.5.6.3 Working Calibration Standards: Store this standard at room temperature for up to one year. Dilute Intermediate Standard solution to make the following concentrations:

Std #1		mL into 50 mL
PCBSA	1 ppm	0.5

Std #2		mL into 40mL
PCBSA	5 ppm	2

Std #3		mL into 50 mL
PCBSA	10 ppm	5

Std #4		mL into 40 mL
PCBSA	50 ppm	20

Std #5		g into 1L
PCBSA	100 ppm	0.1

*same as the Int. Std.

9.5.7 ICV/LCS/MS Standards: The LCS and MS source are used from a non-calibration source.

9.5.7.1 Lab Control Spike: The standards below for Cl, NO_3, and SO_4 are made from 1000 ppm stock solution into a graduated cylinder and brought up to volume of 100 mL with nanopure. Solution is prepared weekly.

		Aliquot
Cl	50ppm	0.5mL of 10,000ppm stock
NO_3	50ppm	5 mL of 1000ppm stock
SO_4	50ppm	0.5mL of 10,000ppm stock

9.5.7.2 Matrix Spikes (Cl, NO_3, SO_4): Add the below aliquots into a 5 mL sample. Adjust spike amount to 1750 uL and final volume to 51.75 mL in Element.

			Aliquot
Cl	High	50ppm	25 uL of 10,000 ppm stock
NO_3	High	20ppm	100 uL of 1000 ppm stock
SO_4	High	100ppm	50 uL of 10,000 ppm stock

9.5.7.3 PCBSA LCS/ICV Standard:

9.5.7.3.1 Stock Standard Salt: 4-Chlorobenzenesulfonic acid purchased from a certified vendor that is from a noncalibration source. Store at room temperature for up to 10 years from date received.

9.5.7.3.2 Intermediate Standard 100 ppm: Weigh 0.1 g of Chlorobenzenesulfonic acid into 1L of nanopure. Store this standard at room temperature for up to one year.

9.5.7.3.3 Working Standard 25 ppm: Dilute 10 mL of the intermediate standard into 40 mL of nanopure.

10.0 Quality Control

Note: See also ESB SOP Q01 for general QC requirements

10.1 Our laboratory has a formal quality control program. The minimum requirements of this program are stipulated below.

10.1.1 In recognition of the rapid advances occurring in chromatography, the analyst is permitted certain options to improve the separations or lower the cost of measurements. Each time such modifications to the method are made the analyst is required to repeat the procedure in Section 10.2.

10.2 Initial Demonstration of Proficiency: Before performing any analyses, the analyst demonstrates the ability to generate acceptable accuracy and precision with this method.

10.2.1 Each analyst will analyze four replicates of a standard at the NELAP level of 1-4x RL. The average percent recovery of the QC samples must be 80-120% with a maximum %RSD of 10.

10.3 Demonstration of Continuing Proficiency: On an annual basis, analyst must turn in valid LCS data from four consecutive LCS's or results from a successful Performance Evaluation Study. LCS percent recovery must meet laboratory prescribed acceptance criteria. Relative standard deviation between the four replicates must be less than or equal to 20%. Performance Evaluation Studies are performed twice a year or more. Quality control check samples are analyzed *twice a year* with the performance evaluation sample studies required to maintain state certification. *Study limits are used to evaluate results.*

10.4 Calibration Blank/Method Blank:

10.4.1 An aliquot of Nanopure water is analyzed at the beginning of each run, every ten samples, and at the end of the run for the calibration blank. One of these blanks is reported for every 20 samples to satisfy the method blank requirement.

10.4.2 Results must be below but not more negative than the *reporting limit*. Sample results at or above the reporting limit must be accompanied with a qualifier if the method blank exceeds a concentration greater than 1/10 the measured concentration of the sample. Blank results below the reporting limit are considered to be ND and will not require a note.

10.5 Lab Control:

10.5.1 In order to verify that standards have been prepared correctly a LCS is performed using a standard of known concentration from an

independent source. This laboratory control sample containing each analyte of concern is analyzed with each batch of samples processed. If more than 20 samples are run in a batch analyze one LCS for every 20 samples and report per matrix type.

10.5.2 Evaluate the accuracy by comparing to laboratory acceptance criteria. Results may be generated historically but must be as tight as 90-110%. The acceptance criterion for PCBSA is 80-120%.

10.5.3 If acceptable data cannot be obtained, locate the problem and correct it. If during the course of a run a LCS is out of range, it is rerun on the spot if possible. If this is not possible the analyst may reevaluate the data based on peak height rather than peak area. If this does not solve the problem a fresh calibration is performed and all samples are rerun starting from the last acceptable LCS.

10.6 Matrix Spike and Matrix Spike Duplicate:

10.6.1 Matrix spikes are performed at a frequency of 10% by analysis of a MS every 10 samples per matrix type. MSD is done per 20 samples.

10.6.2 Historical limits are used for percent recovery requirements with a maximum RPD of 20%. Limits should be 80-120% or better. If the concentration of fortification is less than 25% of the background concentration of the sample, the matrix recovery should not be calculated and a qualifier (QM4x) is attached.

10.7 Replicates are analyzed for solid samples. The maximum allowable RPD is 40%.

10.8 When doubt exists over the identification of a peak in the chromatogram, confirmatory techniques such as sample dilution and fortification must be used.

10.9 Linear Calibration Range: *This method uses an eight point calibration curve so that if any portion of the range is shown to be nonlinear, that portion is clearly defined. Results are not accepted over the high standard value.*

10.10 An MDL study is completed whenever major equipment or procedural changes are made. Standards are spiked at the reporting limit or 2-3 times the estimated instrument detection level. A minimum of seven replicates is analyzed. See QA Manual for calculation. Results must be below the reporting limit. The MDL of the least sensitive instrument is entered into LIMS.

10.10.1 LOD Verification: On a yearly basis, an aliquot of water is spiked at the level of 2-3 times the MDL. The sample is analyzed.

Analyte response must be greater than method blank response to verify the MDL for each column.

11.0 Procedure

11.1 Set-up:

11.1.1 Prepare Eluant. Turn Air valve to 3psi. Check that the air line is connected to the eluant bottle. Set pump rate at 1.2 mL/min.

11.1.2 On Chromeleon Software program – click on run icon. Under file click on load method – anion 300.

11.1.3 Wait for conductivity and pressure to stabilize.

11.2 Standardization and Calibration:

11.2.1 Using a clean syringe, fill one vial with the Method Blank by filtering Nanopure through a 0.2 µm disc filter.

11.2.1.1 Place vial in position #1 of autosampler.

11.2.1.2 Start autosampler per manufacturer's instructions

11.2.2 Using a clean syringe, fill one vial with an initial calibration verification standard.

11.2.2.1 Place vial in position #2 of autosampler.

11.2.2.2 Start autosampler per manufacturer's instructions

11.2.3 The initial calibration verification standard should read within the established control limits. If it does not, re-inject it, if it still does not work, recalibrate.

11.2.3.1 Load calibration standards on the autosampler.

11.2.3.2 Inject eight calibration standards.

11.2.4 Check an initial calibration verification standard again.

11.3 Analysis:

11.3.1 Fill vials with sample. Shake sample and if turbid, filter through a 0.2 µm disc filter.

11.3.1.1 Start autosampler per manufacturer's instructions

11.3.2 Make sure the Chromeleon software is calculating appropriately by observing peak heights/areas and retention times.

11.4 Shutdown

11.4.1 Under Run – load stop method.

11.4.2 Turn pressure valve to 0 psi.

11.5 Check system calibration daily and, if required, recalibrate as described in Section 9.

11.6 The injection loop is flushed thoroughly, using each new sample. The same size loop is used for standards and samples. Record the resulting peak size in area or peak height units.

11.7 The width of the retention time window used to make identifications is based on a study of measurements of actual retention time variations over the course of a day. Three times the standard deviation of a retention time is used to calculate the window size unless experience shows that the window requires adjustment. The experience of the analyst weighs heavily in the interpretation of chromatograms. The current window is $\pm$ 0.5 minutes for chloride and nitrate and $\pm$ 1 minute for sulfate.

11.8 If a sample analyte concentration exceeds the calibration range, the sample is diluted to fall within the range. Samples with reportable results $\leq$ 10 X the reporting limit are reanalyzed if proceeded by a sample that exceeds the concentration of the highest calibration standard noted below:

Cl	NO_3	SO_4 mg/L
250	100	250

11.9 If the resulting chromatogram fails to produce adequate resolution, or if identification of specific anions is questionable, fortify the sample with an appropriate amount of standard and reanalyze.

11.10 Wherever possible evaluate co-eluding peaks by redrawing the baseline rather than sample dilution and document on your raw data.

11.11 Solid Sample Preparation: The following extraction should be used for solid materials. Add an amount of reagent water equal to ten times the weight of solid material taken as a sample. This mixture is agitated for sixty minutes by shaking intermittently. Filter the resulting slurry before injecting using a *0.20* micron membrane type filter. This can be the type that attaches directly to the end of the syringe. *A sample is duplicated for every batch prepared*. Add N_WEX to Solid/sludges and N_SAG to soils.

11.12 When manual integration is performed, ensure that it is obvious this has occurred. It must be documented and traceable in both the electronic data and the hard-copy printouts. Copies of both the original and modified chromatogram are kept with the run. If the manual integration required to

achieve proper peak shape does not affect the result by more than 5% than the original integration may still be used.

12.0 Calculation

12.1 Prepare separate calibration curves for each anion of interest by plotting peak size in area, or peak height units of standards against concentration values. The system will then compute sample concentration by comparing sample peak response with the standard curve.

12.2 Report results in mg/L.

12.3 Report:
NO_2- as N
NO_3- as N or as NO_3 if desired by the client
$H(PO_4)_2$- as P

12.4 Dilutions:
Raw results are displayed on the chromatograms. Dilutions are noted on the schedule and entered into LIMS by editing the primary prep volume.

12.5 The "operator" noted on the run is the chemist who filtered and loaded the samples. The chemist who, prior to entry: examines the run, evaluates peaks, performs manual integrations, calculates QC results, reprocesses calibrations, or determines the need for reanalysis is the person who is entered into LIMS as the analyst.

12.6 Prepared time is entered as the date and time the sample was analyzed, since there is no preparation step. Sample analysis time is assigned individually by the instrument upon data acquisition.

12.7 Calculations associated with this method:

12.7.1 Total Anions (TA)

mequiv. of OH + CO3 + HCO3 + SO4 + Cl + NO3 = TA

12.7.2 Electrochemical Balance (ECB)

Total Cations (TC) – Total Anions (TA)

12.7.3 Total Dissolved Solids by Summation (TDSSUM)

mg/L of 0.6(Total Alkalinity) + Na + K + Ca + Mg + SO4 + Cl + NO3 + F + SiO3 = TDSSUM

13.0 Reporting: ESB Notification Procedure of Nitrate Violations

13.1 Analyst analyzes sample

13.2 Results are quantified

13.3 Confirmation

- 13.3.1 Drinking water Nitrates that are ≥ 45ppm (MCL) are first checked for accurate ID (compare LIMS label to client label) and then re-injected if they are not historically expected. Result confirms if the RPD of the two results ≤ 10.
- 13.3.2 If result does not confirm within the RPD criteria, inject sample a third time to determine the true result.
- 13.3.3 Discuss **challenging chromatography** issues with the Lab Director before PM notification. A result above the MCL is very costly to the client.

13.4 Once the result is confirmed by quantification of re-injection, the 24hr* clock is ticking.

13.5 Enter the **higher** of the two results in LIMS with a qualifier (Nconf). Place the confirmation result in the "Analyte Info1" column.

13.6 Notify the PM by placing the **final** result and duplicate result (or H for historically high nitrate) on the bench sheet. Also note any challenging chromatography issues. Place date and time of quantification of the re-injection on the bench sheet.

13.7 The PM must contact the client using the emergency contact list. If the PM is unable to **speak to a person** within a time period, they must notify the State of the MCL exceedance before the time period expires.

- 13.7.1 Inform the client of the limited time frame in which to resample and reanalyze, and of a potential rush charge.
- 13.7.2 Tell them to notify Login personnel upon submission of a "short holding time resample."
- 13.7.3 Place notification date and time on the bench sheet. Note if client will not be resampling. Return bench sheet to IC Lab until resample is completed.

13.8 When a resample is submitted to the laboratory, the analyst must be notified so it can be analyzed within the 24hr** time frame.

- 13.8.1 If the resample confirms the original result it is injected only once.
- 13.8.2 The analyst places the final result on the new bench sheet from the original sample and gives to the PM.
- 13.8.3 Once the client or state is notified (see section 13.7) the bench sheets are stored by month.

Regulatory Time Frame	**Nitrate**
Analyze sample within holding time.	48hrs

Maximum Level before client is out of compliance.	45.4ppm
Lab notifies client once result is "obtained" (quantified/confirmed).	24hrs*
Client must resample and reanalyze once notified by lab.	24hrs**
Lab notifies client once resample result is "obtained."	24hrs
If average of two results exceeds MCL, client notifies State.	24hrs

14.0 Transferring Data

14.1 Update the Schedule/Run Log. Analyst will usually make notes on the printed schedule the night before for such things as dilution changes, sample bottle letters, and referenced samples for spikes. These things are not generally typed in initially because they are not always known for sure when they start loading the ICs.

14.2 Reference Batch QC on Bench Sheet. (Element\Laboratory\Bench Sheet\Batch#) Source sample(s) for MS/MSD(s) are referenced. Dilutions are entered by editing the Initial column with the appropriate amounts for a Final amount of 50 mL (for solids – any dilution on top of the normal 1:10 extraction is entered in the Diln column of the Data Entry/Review screen). Spike Amounts and Spike IDs are entered.

14.3 Create a Data Entry Table. (Element\Laboratory\Data Entry/Review\Batch#) Select the analytes associated with the batch, click Create, click Export and select an Elmnt User File to save the batch in (ex. IC-WATERTEMP), click Save and then click Cancel back in Data Entry/Review.

14.4 Process/Transfer Batch. Select the Input tab. Click Select to acquire a complete schedule of samples to send or click Build to select individual data files (if Build is selected, the file will need to be saved as a new file name – do not save as an existing file name). Also in the Input tab under Process Methods click the circle next to From Schedule. Once you have selected or created a schedule click the Export tab and there click Browse to select a file name (ex. WATERS) in which to save the data to send to Data Tool and click Save. Also in the Export tab select a report type by clicking the circle next to Full report type. When finished click OK at the bottom of the Processing window. Then click the Start icon (looks like an arrow circling clockwise) or under the Processing menu select Start. This sends the data to Data Tool.

14.5 Merge Files in Element DataTool and make edits. (Element\Laboratory\Data Tool Main) In Data Tool under Element Data Entry Table click Browse. Select the Element User File that was created above (ex. IC-WATERTEMP) and click Open. Under Instrument Data Files click Browse. Click Clear to remove previous work. From the bottom right box double click the file exported from Batch (ex. WATER), the samples will appear in the top left

box. Highlight the desired samples, click Include, and then click Done. In Data Tool Main click Merge Files.

14.6 Make Necessary edits. In Data Transfer select the Instrument Data tab. Do a print screen of samples and paste on to a Word page and print each page (these pages are useful for documenting any needed edits or problems with QC). Upon reviewing chromatograms and noting any edits or problems, make any sample name or result edits to the appropriate lines in the Instrument Data table. Click Refresh. Select the Merged Upload tab. Remove the lines of samples that should not be entered (ex. need dilution, associated with bad CCV, etc.). When all edits have been made and lines removed click Save. Save as the User File from above (ex. IC-WATERTEMP).

14.7 Enter, Save, and QC review. (Element\Laboratory\Data Entry/Review) In Data Entry/Review click Open and double click the saved User File (ex. IC-WATERTEMP). Click Save. Click Query for each analyte to see if there are any red lines (QC problems, holding time issues, etc.). Make any necessary edits. Add appropriate qualifiers where needed.

15.0 Corrective Action For Out of Control Or Unacceptable Data:

See SOP Q06 – Corrective Action

16.0 Pollution Prevention and Waste Management:

16.1 Instrument Waste is dumped in the sink and neutralized.

16.2 SOP S05 – Neutralization Procedure for Acid and Alkaline Wastes

SOP S07 – Pollution Prevention

17.0 Method Performance

See Method 300 Tables 3-9.

Our intralaboratory generated data is expected to achieve similar results. Refer to Refer to MDL studies, Initial Demonstration of Capability Studies, and laboratory control charts maintained in the QA Office.

References:

EPA SW846 Method 9056

EPA Methods for the Determination of Inorganic Substances in Environmental Samples, (August 1993) Method 300.0 revision 2.1

EPA Method ML300.0 Determination of Inorganic Anions by Ion Chromatography, Revision 1.0 November 2007

California Department of Health Services IC Rev 0

Note: All *italicized items* are an indication of a variation from the method.

Approved by Stacey A. Fry Date 04/02/2014

Nitrogen, Nitrite (Spectrophotometric)
Babcock Laboratories, Inc.
Standard Operating Procedure
SM 4500-NO_2 B
Date Effective: 6/24/14

1.0 Scope and Application

1.1 This method is applicable to the determination of nitrite in drinking, surface and saline waters, domestic and industrial wastes. This method may also be used for water extractions of solid materials if both client and regulator agree to this variation.

1.2 The method is applicable in the range from 0.01 to 1.0 mg NO2-N/L as proven by a yearly curve. The reporting limit is 0.10 mg NO2-N/L.

1.3 Environmental Relevance:

1.3.1 Contamination is caused by runoff from fertilizer use, leaching from septic tanks, sewage, and erosion of natural deposits. Nitrites are very soluble and do not bind to soils. They are likely to remain in water until consumed by plants or other organisms.

1.3.2 Nitrite-N has a MCL of 1 mg/L and is the etiologic agent methemoglobinemia.

2.0 Summary of Method

2.1 The diazonium compound formed by diazotation of sulfanilamide by nitrite in water under acid conditions is coupled with N-(1naphthyl)ethylenediamine dihydrochloride to produce a reddish-purple color which is read in a spectrophotometer at 543 nm.

3.0 Sample Handling and Preservation

3.1 Samples should be analyzed as soon as possible. They may be stored for up to 48 hours at 4^{o}C. Never use acid preserved samples.

4.0 Interferences

4.1 There are very few known interferences at concentrations less than 1,000 times that of the nitrite; however, the presence of strong oxidants or reductants in the samples will readily affect the nitrite concentrations.

4.2 High alkalinity (>6000 mg/L) will give low results due to a shift in pH. The ideal pH for nitrite analysis is 5-9.

4.3 NCl3 imparts a false red color when color reagent is added.

4.4 The following ions cause precipitation: Sb^{3+}, Au^{3+}, Bi^{3+}, Fe^{3+}, Pb^{2+}, Hg^{2+}, Ag^{+}, $PtCl_6^{2-}$, and VO_3^{2-}.

4.5 Cupric ion may cause low results by catalyzing decomposition of the diazonium salt.

4.6 Chlorine converts NO2 and may be the reason for low results.

4.7 Suspended material is removed by filtration. Color is removed by filtration or by subtracting a background absorbance reading.

5.0 Apparatus

5.1 Filtration apparatus or 0.45 micron disk filters.

5.2 Genesys 10 Spectrophotometer (#227) equipped with 1 cm or larger cells for use at 543 nm.

5.3 Standard laboratory glassware: volumetric flasks, 100 mL beakers, graduated cylinders, glass pipets, autopipet.

Note: All glassware is cleaned immediately after use by thorough rinsing with three portions of D.I. water. If glassware still appears dirty, further steps are taken, by use of one of the following: Alconox and hot water, 1:1 acid rinse, acetone or appropriate solvent rinse. Glassware is always finished with a final D.I. rinse.

6.0 Reagents and Standards

6.1 Reagent water: D.I. water free of nitrite and nitrate is to be used in preparation of all reagents and standards and for sample dilutions.

6.2 Color Reagent:
- 6.2.1 To 800 mL of D.I. add 100 mL 85% phosphoric acid and 10 grams of sulfanilamide.
- 6.2.2 Mix until sulfanilamide is dissolved completely.
- 6.2.3 Add 1 gram N-(1 naphthyl)-ethylenediamine dihydrochloride, mix to dissolve.
- 6.2.4 Dilute to 1 L with D.I. water.
- 6.2.5 Solution is stable for about a month when stored in a dark bottle in the refrigerator.

6.3 Stock Standard: Solution is stored in the walk-in refrigerator until manufacturer specified holding dates. Once opened, the expiration date is 1 year from date opened as long as it does not exceed manufacturer's original expiration date. Stock standards can be replaced sooner if comparisons with QC samples indicate a problem.

- 6.3.1 Stock #1: Purchased 1000 ppm certified sodium nitrite standard solution. Concentration of actual nitrite in this solution is calculated to be **304 ppm.**
- 6.3.2 Stock #2: Purchased 1000 ppm certified nitrite-nitrogen standard solution.

6.4 Working Standard: These solutions are made fresh with each use in D.I. water. Mix standards well. Invert and shake several times.
- 6.4.1 Midpoint Check Standard Solution (0.12 mg/L):
  - 6.4.1.1 Dilute 1 mL of stock standard #1 to 500 mL in a volumetric flask.
  - 6.4.1.2 Transfer 10 mL of the above solution into the beaker and dilute to 50 mL.
- 6.4.2 Lab Control Standard Solution (0.1 mg/L):
  - 6.4.2.1 Dilute 0.25 mL of stock standard #2 to 500 mL in a volumetric flask.
  - 6.4.2.2 Use 10 mL in the beaker and dilute to 50 mL.
- 6.4.3 Calibration Standards:
  - 6.4.3.1 When a new curve is necessary, stock standard #1 is diluted to the necessary concentrations. See Section 8.0 for recipe of calibration standards.

7.0 Procedure

7.1 Sample Set Up – Check pH of samples and adjust if not between 5 and 9. If samples are filtered, all QC samples need to be filtered through the same process. See below for more information.

- 7.1.1 Use 3 beakers: one for the blank, midpoint check and lab control.
  - 7.1.1.1 Method Blank: The method blank is *used to tare the instrument.* Add 50 mL of D.I. water to the first beaker for the reagent blank. If samples are filtered, filter 5mL of D.I. water and add 45 mL of unfiltered D.I. to bring to a final volume of 50 mL.
  - 7.1.1.2 LCS: Add 40 mL of D.I. water and 10 mL of LCS Standard Solution to the second beaker. If samples are filtered, filter 5 mL of D.I. water. Add 35 mL of unfiltered D.I., and 10 mL of LCS standard to bring to a final volume of 50 mL.
  - 7.1.1.3 MPC: Add 40 mL of D.I. water and 10 mL of Midpoint Check Standard to the last beaker in the batch. If samples are filtered, filter 5 mL of D.I. water. Add 35 mL of unfiltered D.I., and 10 mL of Midpoint Check Standard to bring to a final volume of 50 mL.
- 7.1.2 Use 2 beakers: one for the MS and MSD samples.
  - 7.1.2.1 Add 5 mL of sample (filter sample if not clear), 35 mL of D.I. water, and 10 mL of LCS standard to bring to a final volume of 50 mL.
- 7.1.3 Set up the rest of the samples in beakers by adding 5 mL of sample and 45 mL of D.I. water.
  - 7.1.3.1 If the sample is not clear, filter it. If filtrate is discolored or turbid, take a background reading. Zero the spec to a blank prior to addition of color reagent. Read the sample prior to addition of color reagent. This absorbance is subtracted from the reading acquired in section 7.5. If results are still out of range, a serial dilution may be necessary.
- 7.1.4 For solid samples: Add an amount of reagent water equal to ten times the weight of solid material taken as a sample. This mixture

is agitated for sixty minutes by shaking intermittently. Filter the resulting slurry through a 0.45 micron membrane type filter. Place 1-5 mL of extract and 45 mL of D.I. water into sample beaker. Document the ratio used on the data page.

7.2 Add 2 mL of the color reagent to each sample. Swirl.

7.3 Let sit for at least 10 minutes for color development.

7.4 Zero the spectrophotometer to the reagent blank.

7.5 Read samples on UV/V Spectrometer at 543 nm. Record the answer in absorbance units.

8.0 Calibration and Calculation

8.1 Three calibration standards are required by the method. In general calibration standards are prepared at six concentrations, including a calibration blank. A calibration curve is analyzed on a yearly basis covering the following concentration range.

8.1.1 The Midpoint Check Stock Standard #1 is used.

8.1.1.1 0.05 mg/L - Add 0.4 mL of MPC standard in 500 mL. Add 10 mL of this solution and dilute up to 50 mL of D.I.

8.1.1.2 0.12 mg/L - 0.12 mg/L - Add 1 mL of MPC standard in 500 mL. Add 10 mL of this solution and dilute up to 50 mL of D.I.

8.1.1.3 0.18 mg/L - Add 1.5 mL of MPC standard in 500 mL. Add 10 mL of this solution and dilute up to 50 mL of D.I.

8.1.1.4 0.24 mgL - Add 2 mL of MPC standard in 500 mL. Add 10 mL of this solution and dilute up to 50 mL of D.I.

8.1.1.5 0.3 mg/L - Add 2.5 mL of MPC standard in 500 mL. Add 10 mL of this solution and dilute up to 50 mL of D.I.

8.2 An acceptable curve yields an $r^2 = 0.99$ or better.

8.3 A regression equation is generated from the curve.

8.4 Results are calculated from the regression equation.

8.5 Enter the raw concentration into the LIMS. Ensure that the proper initial volume is entered on the bench sheet.

8.6 Immediately following calibration a 0.1 mg/L ICV is analyzed using the LCS source to verify the curve. Results must be within 90-110% of the expected value or a new curve may be necessary.

8.7 Continuing Calibration Verification Checks: The calibration is verified on a daily basis by analysis of the Lab Control, at the beginning of the day's batch, and the Midpoint Check at the end of the day's batch. See acceptance criteria below.

8.8 Results greater than the highest calibrator must not be reported. Dilute result into the calibration range.

9.0 Quality Control:

Note: See also ESB SOP Q01 for general QC requirements

9.1 A LCS is analyzed once per batch or 1 for every 20 samples, whichever is greater and reported per matrix type. Compare the Laboratory Control results to the acceptance ranges. If the results are not within the acceptance ranges of 90% - 110%, the analysis is considered to be out of control and the problem must be corrected before the analysis proceeds.

9.2 Matrix Spikes (MS and MSD) are analyzed with each batch of 20 or fewer samples per matrix type. Calculate the percent recoveries of the spikes by the following equation:

(Spiked sample - sample) /1 X 100

If the spike recovery does not fall within acceptance ranges of 80%-120% max RPD of 20% for aqueous samples and historically generated limits for solid samples, the spike must be re-analyzed. If the second spike still does not fall within acceptance ranges, the client's results are flagged.

9.3 The Midpoint Check is analyzed with each batch of 20 or fewer samples. It must fall within the same criteria as the lab control, 90-110%, or the validity of the curve is in question. If the midpoint check is remade and it is still out of range, a new curve may have to be drawn.

9.4 A standard at the reporting limit is analyzed at the end of the run. This is used to validate samples reported as none detected and to evaluate instrument sensitivity should calibration checks show a low bias. This standard should have a signal greater than the method blank.

9.5 An MDL study is completed whenever major equipment or procedural changes are made. Standards are spiked at the reporting limit or 2.5-5 times the estimated MDL and a minimum of seven consecutive replicates is analyzed. See QA Manual for calculation. Results must be below the reporting limit.

9.6 LOD Verification: On a yearly basis, a QC sample is spiked at a level of not more than 2-3 times analyte MDL. The sample is analyzed. Analyte response must be greater than the method blank response to verify the Level of Detection or MDL.

9.7 Initial Demonstration of Capability: Prior to analysis of samples or when a significant change is made to the method, an Initial Demonstration of Capability Study is performed. This is accomplished by analysis of four replicates of a QC sample made at a concentration 10 times the MDL to

midrange of the curve. The average percent recovery of the QC samples must be 80-120% with a maximum %RSD of 20.

9.8 Demonstration of Continuing Proficiency: On an annual basis, each analyst must turn in valid LCS data from four consecutive LCS samples or results from a successful Performance Evaluation Study. LCS percent recovery must meet laboratory prescribed acceptance criteria. Relative standard deviation between the four replicates must be less than or equal to 20%. Performance Evaluation Studies are performed twice a year.

10.0 Reporting

10.1 Results are reported as mg/L Nitrite-N for liquids and in ug/L for waters.

10.2 The reporting limit is 0.1 mg/L (100 ug/L). Refer to Element for MDL information.

10.3 Analysis time is documented as the time the absorbance of the last sample in the batch was read.

10.4 Set the prepared time to equal the analyzed time in Element.

10.5 The results are reported to 2 significant figures.

11.0 Definitions: See SOP Q15 – SOP Definitions

12.0 Safety:
See SOP S01 – Concentrated Acids and Bases
SOP S03 – Spill Control Policy

13.0 Corrective Action For Out of Control Or Unacceptable Data:
See SOP Q06 – Corrective Action

14.0 Pollution Prevention and Waste Management:
See SOP S05 – Neutralization Procedure for Acid and Alkaline Wastes
SOP S06 – Disposal of Chlorinated Solvents
SOP S07 – Pollution Prevention

15.0 Method Performance:
Refer to MDL studies, Initial Demonstration of Capability Studies, and laboratory control charts maintained in the QA Office.

16.0 References/Method Source:

EPA 354.1 Methods for Chemical Analysis of Water and Wastes.

Standard Methods for the Examination of Water and Wastewater: APHA/AWWA/WEF, 22nd edition. 4500-NO_2 B

GENESYS 10 Operator's manual Thermo Electron Corporation

Note: All *italicized items* are an indication of a variation from the method.

Approved by Julia Sudds Date 6/17/14

ORTHO-PHOSPHATE PHOSPHORUS
Babcock Laboratories, Inc.
STANDARD OPERATING PROCEDURE
ASCORBIC ACID METHOD
(SM 4500-P E)
Effective Date: __04/07/14___

1.0 Scope and Application

This method covers the determination of ortho phosphate phosphorus in drinking, surface and saline waters, domestic and industrial wastes.

1.1 Health Effects of Phosphorus

1.1.1 Too much phosphate can cause health problems, such as kidney damage and osteoporosis.

1.2 Environmental Effects of Phosphorus

1.2.1 In soil, phosphorus will remain for several days, but in deep soils and the bottom of rivers and lakes phosphorus will remain for a thousand years or so. Orthophosphates applied to agricultural or residential cultivated land as fertilizers are carried into surface waters with storm runoff.

1.2.2 The increasing phosphorus concentrations in surface waters, raises the growth of phosphate-dependent organisms, such as algae and duckweed. These organisms use great amounts of oxygen and prevent sunlight from entering the water. This is known as eutrophication.

2.0 Working Range: 0.05 – 1.0 mg/L

3.0 Summary

Ammonium molybdate and antimony potassium tartrate reacts in an acid medium with dilute solutions of phosphorus to form an antimony-phospho-molybdate complex. This complex is reduced to an intensely blue-colored complex by ascorbic acid. The color is proportional to the phosphorus concentration. Only orthophosphate forms a blue color in this test.

4.0 Sampling and Preservation

4.1 Samples must be unpreserved and stored in the refrigerator.

4.2 Analysis must be done within 48 hours. *Samples are filtered if necessary prior to analysis.*

5.0 Interferences

5.1 Copper, iron, or silicate at concentrations many times greater than their reported concentration in seawater causes no interference. However, high iron concentrations can cause precipitation of and subsequent loss of phosphorus.

5.2 Arsenate is determined similarly to phosphorus reacting with molybdate reagent to produce a blue color and should be considered when present in concentrations higher than phosphorus. Concentrations as low as 0.1 mg As/L can interfere.

5.3 Hexavalent chromium and nitrite interfere to give results about 3% lower at concentrations of 1 mg/L and 10-15% lower at 10 mg/L.

6.0 Safety

General laboratory safety procedures are sufficient for this analysis. Recommended safety equipment includes gloves and safety glasses.

7.0 Equipment

7.1 Standard laboratory glassware: volumetric flasks, beakers, graduated cylinders, pipettes.

7.2 Syringe and 0.45 micron filter disk

7.3 Spectrophotometer: Spectronic Genesys 10 uv

8.0 Glassware Preparation

8.1 All glassware is acid washed in 1:1 HCl immediately after use, followed by thorough rinsing with three portions of D.I. water. If glassware still appears dirty, further steps are taken, by use of one of the following: Alconox and hot water, 1:1 acid rinse, acetone or appropriate solvent rinse. Glassware is always finished with a final D.I. rinse.

8.2 Glassware is stored separately for Ortho-Phosphate Phosphorus use only.

9.0 Phosphate Reagents

9.1 Reagent water: Nanopure

9.2 P-Reagent - Let all reagents reach room temperature before mixing. Mix in the order given. If turbidity forms in the combined reagent, shake and let stand for a few minutes until turbidity disappears before proceeding. The reagent is stable for 4 hours at room temperature. *If refrigerated, the reagent can be kept and used for 3 days assuming all QC samples meet the proper acceptance criteria. Usually this reagent is kept no longer than 2 days.* Place reagent immediately back in the refrigerator after use to preserve its integrity. When fresh the reagent should be light yellow. If the reagent turns gold, it is too old. Store reagent in glass.

9.2.1 Prepare two beakers as follows:

9.2.1.1 Beaker #1 - 0.53 g Ascorbic acid and 30 mL nanopure water. Make fresh each time.

9.2.1.2 Beaker #2 – 50 mL 5N H_2SO_4, 5 mL Potassium Antimonyl Tartrate solution, and 15 mL Ammonium Molybdate solution.

9.2.2 Add the contents of beaker #1 to beaker #2. This will make 100 mL P Reagent.

9.2.3 5N Sulfuric Acid: Start with 430 mL of nanopure water and slowly, with stirring, add 70 mL of concentrated H_2SO_4. Let cool and dilute to 500 mL. Reagent is stored in a glass bottle at room temperature for up to one year.

9.2.4 Ammonium Molybdate solution: Dissolve 20 g of Ammonium Molybdate 4-hydrate $[(NH_4)_6Mo_7O_{24}.4H_2O]$ in 500 mL nanopure water. Reagent is stored in a glass bottle at room temperature for up to three months.

9.2.5 Potassium Antimonyl Tartrate Hemihydrate: Dissolve 1.3715 g $K(SbO)C_4H_4O_6 \cdot 1/2H_2O$ in 400 mL nanopure water. Dilute to 500 mL. Reagent is stored in a glass bottle at room temperature for up to six months.

9.3 Calibration Standards for Curve:

9.3.1 Stock: KH_2PO_4 previously dried in a 105^oC oven and stored in a desiccator.

9.3.2 Intermediate Calibration Standard (1000 ppm): Weigh up 4.394 g KH_2PO_4 into 1 L of Nanopure. Keep at room temperature. This standard may be kept for up to 12 months or replaced sooner if comparison with QC samples indicates a problem.

9.3.3 Calibration Standards:

9.3.3.1 Intermediate (1 ppm): Pipette 1 mL of 1000 ppm PO4-P Intermediate standard into 1L of Nanopure water.

9.3.3.2 Dilute the 1 ppm Intermediate standard to make a 6 point curve to the following concentrations: 0.05 ppm, 0.1 ppm, 0.2 ppm, 0.4 ppm, 0.7 ppm, and 1.0 ppm. Solutions are made fresh each time.

9.4 Laboratory Control (0.5 ppm):

9.4.1 Stock: Non-calibration source of KH_2PO_4 previously dried in a 105^{o}C oven and stored in a desiccator.

9.4.2 LCS Intermediate (500 ppm): Dilute 2.197 g KH_2PO_4 into 1 L of Nanopure. This standard may be kept for up to 12 months or replaced sooner if comparison with QC samples indicates a problem.

9.4.3 LCS Working Standard (0.5 ppm): Spike 10 uL of LCS Intermediate into 10 mL of Nanopure water. *Filter* and analyze as specified in section 12.0.

9.4.4 Calibration Check 0.25 ppm: Spike 5 uL of LCS Intermediate into 10 mL of Nanopure water. Analyze as specified in sections 12.4 - 12.6.

10.0 Quality Control

Note: See also ESB SOP Q01 for general QC requirements

10.1 A LCS is analyzed once per batch, or 1 for every 20 samples, whichever is greater and reported per matrix type. The result must be within acceptance ranges of 90-110% or the analysis is considered to be out of control. The problem must be corrected before the analysis can continue.

10.2 Matrix Spike and Matrix Spike Duplicate 0.5 ppm:

10.2.1 Spike 10 uL of LCS Intermediate into 10 mL of sample. Analyze as specified in sections 12.4 - 12.6.

10.2.2 An MS/MSD is analyzed one per batch, or for every 20 samples per matrix type, whichever is greater.

10.2.3 If the matrix spike and matrix spike duplicate is not within the control limits of 80-120%, maximum RPD of 20%, the spikes should be re-analyzed. If the results are the same, the sample used as the spike must be qualified.

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10.3 An MDL study is completed whenever major equipment or procedural changes are made. Standards are spiked at the reporting limit or 2.5-5 times the estimated MDL and a minimum of seven replicates is analyzed. See QA Manual for calculation. Results must be below the reporting limit.

10.4.1 LOD Verification: On a yearly basis, a vial of reagent water is spiked at a level of at least 2-3 times analyte MDL. The sample is analyzed. Analyte response must be greater than method blank response to verify the Level of Detection or MDL.

10.4 Initial Demonstration of Capability: Prior to analysis of samples or when a significant change is made to the method, an Initial Demonstration of Capability Study is performed. This is accomplished by analysis of four replicates of a QC sample made at a concentration 10 times the MDL to midrange of the curve. The average percent recovery of the QC samples must be 80-120% with a maximum %RSD of 20.

10.5 Demonstration of Continuing Proficiency: On an annual basis, analyst must turn in valid LCS data from four consecutive LCS samples or results from a successful Performance Evaluation Study. LCS percent recovery must meet laboratory prescribed acceptance criteria. Relative standard deviation between the four replicates must be less than or equal to 20%. Performance Evaluation Studies are performed twice a year.

11.0 Calibration:

11.1 A six-point curve plus the zero point, required by the method, is analyzed on a yearly basis covering the referenced concentration range.

11.2 An acceptable curve yields an $r^2 = 0.99$ or better.

11.3 A regression equation is generated from the curve.

11.4 Results are calculated from the regression equation.

11.5 Immediately following calibration a 0.5 mg/L ICV is analyzed using the LCS source to verify the curve. Results must be within 90-110% of the expected value or a new curve may be necessary.

11.6 Continuing Calibration Verification Checks: The calibration is verified on a daily basis by analysis of the Lab Control Sample and the 0.25 mg/L Calibration Check analyzed at the end of the day's batch. Recovery must be within 90-110% of expected value.

11.7 A standard at the reporting limit is analyzed at the end of the run. This is used to validate samples reported as none detected and to evaluate instrument sensitivity should calibration checks show a low bias. This standard should have a signal greater than the method blank.

12.0 Procedure for Ortho-Phosphorus

12.1 Adjust the pH of the sample prior to measurement *if it is suspected* to be <6.5 or >8.5. Pour up to 10 mL of sample into beaker. The sample must be clear. Filter through a 0.45 micron disk filter if necessary.

12.1.1 If the filtrate is still turbid or discolored read the background. Zero the spec to a blank *prior to addition of reagents.* Read the sample *prior to addition of reagents.* This absorbance is subtracted from the reading acquired in section 13.2.

12.2 If using less than 10 mL of sample, bring up the volume to 10 ml with nanopure water. Record the volume of sample used.

12.3 Spike 10 uL of LCS Intermediate into the lab control, matrix spike, and matrix spike duplicate.

12.4 Add 1.6 mL of "P-Reagent" to all samples.

12.5 Swirl samples then let them stand at least 10 minutes. Read absorbance after 10 minutes but not more than 30 minutes.

12.6 If the sample is a solid:

12.6.1 Add deionized water in a 1:10 ratio with the soil (i.e. 5 g of sample to 50 mL water.) or any ratio that is convenient.

12.6.2 Allow leaching for 1 hour with frequent stirring or tumble for 1 hour.

12.6.3 Centrifuge extract and filter prior to analysis.

12.6.4 Follow the procedure in section 12.1 – 12.5.

13.0 Reading Samples

13.1 Set Spectrophotometer wavelength to 880 nm.

13.2 Before reading samples on the spectrophotometer, zero instrument on the following blank.

13.2.1 Blank: 10 mL of nanopure water to which 1.6 mL of "P-Reagent" have been added.

13.3 Read samples one at a time, rinsing, blotting and wiping the cuvette in between each one. Read absorbance and record.

14.0 Calculations

14.1 Compare each sample absorbance to a current graph. Convert Absorbance to concentration by plugging spectrophotometer reading into a regression equation. Do not report results higher than the highest calibrator.

14.2 Enter raw results into the LIMS.

14.3 Multiply concentration by Dilution Factor if 10 mL were not used.

14.4 Results are reported in mg/L to 2 significant figures.

14.5 The reporting limit is 0.05 mg/L. Refer to Element for MDL information.

14.6 Analysis time is documented as the time that the absorbance of the last sample in the batch was read.

14.7 Set the prepared time to equal the analyzed time in Element.

14.8 All results are reported as ortho-phosphate phosphorus. If the sample is filtered, no distinction is made for “dissolved” versus “total” since 40 CFR, part 136 Table II instructs all ortho-phosphates to be filtered immediately.

15.0 Definitions: See SOP Q15 – SOP Definitions

16.0 Corrective Action For Out of Control or Unacceptable Data:

16.1 See SOP Q06 – Corrective Action

17.0 Pollution Prevention and Waste Management:

17.1 See SOP S05 – Neutralization Procedure for Acid and Alkaline Waste

17.2 SOP S07 – Pollution Prevention

17.3 Sample waste is placed in a stoppered sink, neutralized and then washed down the drain.

18.0 Method Performance

18.1 Refer to MDL studies, Initial Demonstration of Capability Studies, and laboratory control charts maintained in the QA Office.

References

Standard Methods for the Examination of Water and Wastewater, AWWA/APHA/WEF, 22nd Edition 4500-P E

GENESYS 10 Operator's manual Thermo Electron Corporation

Note: All *italicized items* are an indication of a variation from the method.

Approved by: Julia Sudds Date: 04/03/14

RESIDUE, TOTAL FILTERABLE (Total Dissolved Solids)
Babcock Laboratories Inc.
STANDARD OPERATING PROCEDURE
(Standard Methods, 2540C)
Effective Date: 6/27/14

1. Scope and Application

1.1. This method is applicable to drinking, surface, and saline waters, domestic and industrial wastes.

1.2. This method may also be used for solid samples if both client and regulator agree to this variation. See section 10.14 for details.

1.3. Environmental Relevance –

1.3.1. TDS are both inorganic and organic in origin. Inorganic sources are minerals such as calcium, magnesium, and bicarbonate from rocks. Organic TDS can come from leaves, industrial waste, and sewage. TDS in a water sample comes from natural sources such as the bedrock and decaying plant material and introduced sources such as waste, urban runoff, and chemicals left over from water treatment processes. A constant level of TDS is essential to the health of aquatic organisms as the density of the TDS determines water flow in and out of cells. Elevated TDS levels are not generally health hazards for humans, but have an effect on the smell and taste of drinking water.

1.3.2. The EPA classified TDS as a Secondary Drinking Water Standard and has set a limit of TDS less than 500 mg/L for drinking water. High concentrations of TDS due to carbonates can lead to scale formation and a bitter taste to the water. If a water source tastes salty and an increase in corrosivity is observed, it may have a high TDS concentration due to chlorides. These effects are not necessarily dangerous but are unpleasant. Scale formation is a nuisance as it can be damaging to water heaters and plumbing.

2. Working Range

2.1. 10 mg/L to 2000 mg/L
2.2. The RL is 10mg/l when 200mls of sample are used
2.3. Refer to Element for current MDL information.

3. Summary of Method

3.1. A well-mixed aliquot of the sample is filtered through a standard glass fiber filter. The filtrate is evaporated and dried at 180^{o}C.

4. Sample Collection, Preservation, and Holding Times

4.1. The sample may be collected in either plastic or glass.

4.2. The sample is chilled to 4(± 2) °C.

4.3. The sample must be analyzed within 7 days from collection as specified in 40 CFR part 136, table II.

5. Definitions

5.1. See SOP Q15 – SOP Definitions

6. Interferences

6.1. Highly mineralized waters containing significant concentrations of calcium, magnesium, chloride and/or sulfate may be hygroscopic and will require prolonged drying, desiccation and rapid weighing.

6.2. Samples containing high concentrations of bicarbonate will require careful and possibly prolonged drying at 180°C to insure that all the bicarbonate is converted to carbonate.

6.3. Too much residue in the evaporating dish will crust over and entrap water that will not be driven off during drying. Total residue in the dish should be limited to about 200 mg.

6.4. Static can build up causing scale readings to fluctuate. This could affect the accuracy of the tare or final weights taken. Fluctuations can be reduced by placing a damp paper towel in a blue boat and leaving it in the scale for a minimum of 15 minutes.

7. Safety

7.1. General laboratory safety procedures apply. Care should be taken when reaching over the hot water bath, handling dishes or reaching into the ovens.

8. Apparatus

8.1. EC tubes and rack

8.2. 500 ml side-arm flask.

8.3. Evaporation dishes for water/wastewater.

8.4. Filtration apparatus: Membrane filter funnel with a Whatman grade 934-AH filter disk (or equivalent).
8.5. Drying oven, for operation at 180°C ± 2°C.

8.6. Vacuum aspirator.

8.7. Desiccator with fresh desiccant. Check on the desiccant to make sure that it is always blue. Replace desiccant when it turns purple.

8.8. Balance # 226 with a sensitivity of 0.1 mg. Calibrated on a yearly basis and checked with 5g and 100g class "S" weights on a daily basis. Readings must be within ± 5mg. If values are not within these limits, notify manager and have balance recalibrated.

8.9. Two 12 Unit steam baths (#69, #76). On each day of use clean off the tops of the water baths to prevent residue buildup. On a monthly basis drain the water bath and scrape out the residue. Try to turn off the bath heat when not in use so that cool water can circulate.

8.10. Standard laboratory glassware: volumetric flasks, beakers, graduated cylinders, pipettes.

Note: All glassware is cleaned immediately prior to and after use by thorough rinsing with three portions of D.I. water. If glassware still appears dirty, further steps are taken, by use of one of the following: Alconox and hot water, 1:1 acid rinse, acetone or appropriate solvent rinse. Glassware is always finished with a final D.I. rinse.

9. Standards and Reagents

9.1. 0.01M (746 mg/L) KCl Lab Control: 0.7456 g KCl predried in 105^{o} oven, dissolved in 1 liter of D.I. water. This solution is stored at room temperature for up to 6 months.

10. Procedure

10.1. Instrument Operating Conditions: The oven temperature is set at 180°C. This temperature is checked daily.
10.2. Prepare dishes by heating at 180°C for a minimum of 1 hour.

10.3. Take hot dishes out of 180°C oven. Cool 15-20 minutes on a counter.

10.4. Cool dishes to room temperature in a desiccator. Weigh dishes on balance for tare weight and record. Place dishes on water bath.

10.5. Place a small amount of sample into a tube for electrical conductivity screening. Samples that are high in salts (TDS) will have an elevated conductivity. Filter the appropriate aliquot of sample to give a result of less than 2000 mg/L prior to dilution factors. Refer to table below for guidance. Samples high in organic content might require dilutions beyond those indicated by their EC reading.

EC	mLs of Sample	Dilution
1-100	200	1
100-1500	100	2
1501-4000	50	4
4001-8000	25	8
8001-20000	10	20
20001-40000	5	40
40001-100000	2	100

10.6. Measure proper aliquot of *well shaken* sample, usually 100 ml into a 100 mL graduated cylinder and filter.

10.7. Some samples may be difficult to filter. In order to maximize sample size, try one of the following. If possible do not use less than 10 ml of sample.

10.7.1. A bigger filtration apparatus may be necessary for samples that are not high in salts but contain a great deal of suspended material. It is recommended that the filter time not exceed 10 minutes.

10.7.2. Change filter papers as often as necessary to filter sample ensuring that each filter is rinsed thoroughly.

10.8. After the sample has filtered through, make sure that the filter is completely dry prior to turning off the pump. *Drips should be at least 5 seconds apart and the filter must be visually dry*. Rinse into the filtrate, the filter paper and cylinder with D.I. water. Pour filtrate into the evaporating dish. Rinse flask with a small amount of D.I. water. Add the rinse to the evaporating dish with the filtrate.

10.9. After each sample rinse all apparatus three times with deionized water.

10.10. Place samples on water bath until they are dry. After samples are dry, wipe down the outside of each dish with a paper towel moistened with DI water prior to placing them in the oven.

10.11. After a minimum of one hour remove dishes from oven. Cool dishes for 15-20 minutes on a counter then cool to room temperature in a desiccator.

10.12. Weigh on a balance. Record the weight.

10.13. Place dishes back in the oven for a minimum of 15 minutes, remove, cool and then weigh again. Record the weight.

10.14. This process is repeated until the weights are constant. Constant weight is defined as; “weight change less than 4% of the previous weight or a change less than 0.5 mg, whichever is less” (basically the last digit in the weight cannot vary by more than 5). .

10.15. Use the last final weight taken for each sample to calculate result.

10.16. To clean dishes: Wash inside and outside of evaporating dishes with a green scrubby, 1:1 HNO_3 if necessary, and at least three portions of D.I. Put dishes back in 180°C oven for a minimum of 1 hour. Cool and place in desiccator for the next set.

10.17. Solid samples may be analyzed by first preparing a 1:10 water extract. Add an amount of reagent water equal to ten times the weight of solid material taken as a sample. This mixture is agitated for sixty minutes by shaking intermittently. Filter the resulting slurry through a 0.45 micron membrane type filter. Add filtrate to TDR dish. Report result in the extract, as mg/L, attaching qualifier N-WXr.

11. Calculation and Reporting

$$\frac{(A-B) \times 1{,}000{,}000}{\text{\# ml of sample used}}$$

A = Weight in grams of dish with residue, and
B = Tare weight in grams of dish.

11.1. Alternatively, you may subtract the actual numbers in the weight readings (without any decimal points) and multiply the difference by the factor of 100/(ml of sample used).

11.2. Results are entered into LIMS by taking the

$$\frac{\text{Difference (from data page) X Dilution Factor (based on a 200mL default volume)}}{2}$$

Note: The dilution factor is placed in the DIL column to account for initial volume differences. The initial and final volumes are set at 200 and 100 mL and are not adjusted by the analyst.

11.3. Reporting Units: mg/L

11.4. Reporting Limit: 10 mg/L for 200 ml aliquot or 20 mg/L for 100 ml aliquot

11.5. The date the sample was analyzed is the date recorded. Analysis time is recorded as the time that the last sample is placed in the dish or on the filter. The date and time the sample was analyzed is entered as the preparation date and time.

12. Quality Control:

Note: See also ESB SOP Q01 for general QC requirements

12.1. Duplicates are analyzed with each batch or every ten samples per matrix type whichever is more. The Relative Percent Difference is calculated and compared to the acceptance range. Sample acceptability however is based on a maximum RPD of 20%. If the RPD does not fall within this acceptance range, the sample is re-analyzed. If the RPD still does not fall within the acceptance range, a note is placed on the client's results.

12.2. Laboratory Control: An aliquot of the KCL standard is analyzed with each batch or one every twenty samples whichever is more and reported per matrix type. The result from this analysis is compared to the control limit (90 - 110% recovery). If the result falls outside the control limit, the analysis is considered to be out of control and all the other results in the analytical run are questionable. Troubleshooting is performed to attempt to determine the cause. If the cause is not determined, or the cause is something that could affect the other samples in the batch, the analyses will be re-run.

12.3. A method blank is analyzed with each batch or one every twenty samples whichever is more and reported per matrix type. 200 mL of D.I. water is filtered exactly like a sample. It must read less than 10 mg/L and greater than –10 mg/L for batch acceptance. Reportable sample results must be accompanied with a qualifier if the method blank exceeds a concentration greater than 1/10 of the measured raw concentration of the sample. However, blank results below the reporting limit are considered to be ND and will not require a note.

12.4. An MDL study is completed whenever major equipment or procedural changes are made. Standards are spiked at 2.5-5 times the estimated MDL and a minimum of seven replicates is analyzed. See QA Manual for calculation. Results must be below the reporting limit.

12.5. Initial Demonstration of Capability: Prior to analysis of samples or when a significant change is made to the method, an Initial Demonstration of Capability Study is performed. This is accomplished by analysis of four Lab Control Samples. The average percent recovery of the QC samples must be 80-120% with a maximum %RSD of 20.

12.6. Demonstration of Continuing Proficiency: On an annual basis, each analyst must turn in valid LCS data from four consecutive LCS samples or results from a successful Performance Evaluation Study. LCS percent recovery must meet laboratory prescribed acceptance criteria. Relative standard deviation between the four replicates must be less than or equal to 20%. Performance Evaluation Studies are performed twice a year.

12.7. LOD Verification: On a yearly basis, a QC sample is spiked at a level of 2-3 times analyte MDL. The sample is analyzed. Analyte response must be greater than method blank response to verify the Level of Detection or MDL.

13. Corrective Action For Out of Control or Unacceptable Data:

13.1. See SOP Q06 – Corrective Action

14. Pollution Prevention and Waste Management:

14.1. See SOP S07 – Pollution Prevention

15. Method Performance

15.1. Refer to MDL studies, Initial Demonstration of Capability Studies, and laboratory control charts maintained in the QA Office.

16. References

16.1. Standard Methods for the Examination of Water and Wastewater, AWWA/APHA/WEF, 22nd edition Method 2540C.

Note: All *italicized items* are an indication of a variation from the method.

Approved by: Julia Sudds Date: 6/19/14

RESIDUE, TOTAL SUSPENDED
Babcock Laboratories Inc.
STANDARD OPERATING PROCEDURE
(SM 2540 D)
Date Effective: __6/27/14___

1.0 Scope and Application:

1.1 This method is applicable to all aqueous samples.

2.0 Working Range:

2.1 The working range is 5mg/L (the reporting limit) to 2000mg/L.

3.0 Method Summary:

3.1 A measured volume of sample is filtered through a pre-tared filter. The residue that remains on this filter after drying in a 105°C oven is considered the suspended solid portion of the sample.

4.0 Sample Collection, Preservation and Holding Time:

4.1 The sample must be unpreserved. It must be stored at 4°C until analysis. Analysis must take place within 7 days of sampling per CFR part 136, Table II.

5.0 Interferences:

5.1 Non-representative particulates such as leaves, rocks, and sticks may be excluded.
5.2 To avoid water entrapment, limit the sample size to that yielding no more than 200 mg residue on the filter. (This would be a final result of 2000 mg/L since we are analyzing 100 mLs of sample.)
5.3 For samples high in dissolved solids thoroughly wash the filter with DI water after the sample has passed through the filter.
5.4 Prolonged filtration times resulting from filter clogging may produce high results owing to increased colloidal materials captured on the clogged filter.

6.0 Apparatus, Standards and Reagents

6.1 Side-arm flask of sufficient capacity for sample size selected.

6.2 Filtration apparatus: Membrane filter funnel with a Whatman grade 934-AH glass fiber filter disk (47mm) with a suitable diameter for the funnel.

6.2.1 Each new lot of filters is tested before use. Prepare three filters by rinsing three times with DI water and heating at 105°C for an hour. Take out, let cool, and record tare weight. Place again in the 105°C oven for an hour. Take out, let cool, and record final weight. The difference between tare and final weight should be within MB criteria. See section 9.2.

6.3 Drying oven, for operation at 105°C ± 2°C.

6.4 Vacuum aspirator. Monitor oil level and fill when necessary.

6.5 Desiccator – Check on the desiccant to make sure that it is always blue. Replace desiccant when it turns purple.

6.6 Balance #58 with a sensitivity of 0.1 mg. Calibrated with 0.1 g, 5g, and 100 g class "S" weights on a daily basis. Calibration must be within 5 mg (for 5g & 100g) and 0.5 mg (for 0.1g). If values are not within these limits, recalibrate the balance.

6.7 *Aluminum pans* to hold glass fiber filters.

6.8 Standard laboratory glassware: volumetric flasks, graduated cylinders, pipettes.

Note: All glassware is cleaned immediately prior to and after use by thorough rinsing with three portions of DI water. If glassware still appears dirty, further steps are taken, by use of one of the following: Alconox and hot water, 1:1 acid rinse, acetone or appropriate solvent rinse. Glassware is always finished with a final DI rinse.

6.9 Stock Standards:

6.9.1 Cellulose – stored at room temperature for up to 10 years from date received.

6.10 Lab Control: 500 mg of cellulose is weighed into a liter of DI water. This solution is kept at room temperature for up to 6 months. Use a 100 mL aliquot.

6.11 Reagent water: DI water.

7.0 Procedure

7.1 Each box of glass fiber filters are prepared by rinsing three times with DI water and then dried to a constant weight. Rinsed filters are placed on a tray and heated at 105°C for a minimum of 1 hour. The tray is then removed and cooled to room temperature and then placed in a desiccator. The tray is then weighed and weight recorded. The tray is put back in the oven for a minimum of 15 minutes then removed, cooled and weighed again. This process is repeated until a constant weight is achieved. Lot #, constant weight and date of preparation are noted on bench sheet.

7.2 Use forceps when handling filters. Place filter on balance and record tare weight. Place the filter onto the filtering apparatus. Wet filter with a small amount of DI to seat it.

7.3 Mix sample well by *shaking* sample bottle. Measure an appropriate volume of sample in a graduated cylinder, normally 100 mL. Use a 200 mL sample volume for method blank and samples expected to contain very minute amounts of suspended material. Upon client request a larger volume of sample up to 1000 mL will be used. Filter through apparatus collecting suspended residue on filter. Rinse cylinder and filter 2 to 3 times with a small amount of DI water. Apply suction for three minutes or *until no visible free liquid is present.*

7.3.1 For samples with a lot of suspended matter, a smaller volume of sample (10 – 50 mL) may be used**. Avoid using less than 5 mL.** If less than 10 mLs must be used, an auto pipette can be used to obtain a representative sample by carefully cutting the tip. Verify the accuracy of the altered tip by using the scale. Make sure the tip is rinsed well.

7.4 Place filters in 105°C for a minimum of one hour.

7.5 Remove filters from oven. Cool filters in an area free of moisture or contamination for 10-15 min. Weigh filters or place in a desiccator until ready to weigh.

7.6 Weigh on a balance. Record the weight. Place filters back in the oven for a minimum of 15 minutes, remove, cool and then weigh again. Record the weight.

7.6.1 This process is repeated until the weights are constant. Constant weight is defined as; “weight change less than 4% of the previous weight or a change less than 0.5 mg, whichever is less” (basically the last digit in the weight cannot vary by more than 5).

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7.7 Use the last final weight taken for each sample to calculate result.

8.0 Calculation and Reporting:

$$\frac{(A-B) \times 1{,}000{,}000}{\text{\# mL of sample used}}$$

Where A = Weight in grams of filter with residue, and

B = Tare weight in grams of filter.

8.1 Alternatively, you may subtract the actual numbers in the weight readings (without any decimal points) and multiply the difference by the factor of 100/(mL of sample used).

8.2 Results are entered into LIMS by taking the

$$\frac{\text{Difference (from data page) X Dilution Factor (based on a 200mL default volume)}}{2}$$

8.3 The reporting limit for this procedure (based on a 200 mL aliquot) is 5 mg/L. Current MDL info can be found in Element.

8.4 Report all results to three significant figures.

8.5 The difference is entered as the initial raw result.

8.6 The dilution factor, based on a 200 mL sample aliquot, is entered into the Dil column in LIMS.

8.7 The analyzed time is documented as the time the last sample in the batch was filtered. This is equal to the time filters are placed in the 105°C oven.

8.8 Set the prepared time to equal the analyzed time in Element

9.0 Quality Control:

Note: See also ESB SOP Q01 for general QC requirements

9.1 Duplicates are run with every batch or one every ten samples per matrix type, whichever is more frequent. The Relative Percent Difference is calculated. If the RPD does not fall within the acceptance limit maximum of *25%*, the sample is re-analyzed. If the RPD still does not fall within the acceptance range, a note is placed on the client's results.

9.2 A method blank is analyzed with every batch of samples, at a minimum of once for every 20 samples and reported per matrix type. Results must be less than 5 mg/L and greater than –5 mg/L for batch acceptance. Sample results at or above the reporting limit must be accompanied by a qualifier if the method

blank exceeds a concentration greater than 1/10 of the measured raw concentration of the sample. Blank results below the RL are considered to be ND and will not require a note.

9.3 A lab control is analyzed with every batch at a minimum of once for every 20 samples and reported per matrix type. Results must be between 90-110% recovery for batch acceptance. If the result falls outside the control limits, the analysis is considered to be out of control and all other results in the analytical run are questionable. Troubleshooting is performed to attempt to determine the cause. If the cause is not determined, or the cause is something that could affect the other samples in the batch, the analyses will be re-run.

9.4 An MDL study is completed whenever major equipment or procedural changes are made. Standards should be spiked at 2.5-5 times the estimated MDL. A minimum of six to seven replicates is analyzed. See QA Manual for calculation. Results must be below the reporting limit.

9.5 LOD Verification: On a yearly basis, a QC sample is spiked at a level of 2-3 times analyte MDL. The sample is analyzed. Analyte response must be greater than method blank response to verify the Level of Detection or MDL.

9.6 Initial Demonstration of Capability: Prior to analysis of samples or when a significant change is made to the method, an Initial Demonstration of Capability Study is performed. This is accomplished by analysis of four consecutive Lab Control Samples. The average percent recovery of the QC samples must be 80-120% with a maximum %RSD of 20.

9.7 Demonstration of Continuing Proficiency: On an annual basis, each analyst must turn in valid LCS data from four consecutive LCS samples or results from a successful Performance Evaluation Study. LCS percent recovery must meet laboratory prescribed acceptance criteria. Relative standard deviation between the four replicates must be less than or equal to 20%. Performance Evaluation Studies are performed twice a year.

10.0 Corrective Action For Out of Control Or Unacceptable Data:

See SOP Q06 – Corrective Action

11.0 Pollution Prevention and Waste Management:

SOP S07 – Pollution Prevention

12.0 Definitions: See SOP Q15 – SOP Definitions

13.0 Safety

13.1 General laboratory safety procedures are sufficient for this analysis. Recommended safety equipment includes gloves and safety glasses.

14.0 Method Performance:

Refer to MDL studies, Initial Demonstration of Capability Studies, and laboratory control charts maintained in the QC Office.

15.0 References

Standard Methods For the Examination of Water and Wastewater 22nd Edition APHA/AWWA/WEF 2540D.

HR SERIES Instruction manual 1596-5A-IE-99.03.26

Note: All *italicized items* are an indication of a variation from the method.

Approved by: ___Julia Sudds _____Date: _____6/19/14_____

TOTAL-PHOSPHORUS
Babcock Laboratories, Inc.
STANDARD OPERATING PROCEDURE
ASCORBIC ACID METHOD
(SM 4500-P 5B, E)
Date Effective: 04/07/14

1.0 Scope and Application

These methods cover the determination of specified forms of phosphorus in drinking, surface and saline waters, domestic and industrial wastes.

1.1 Health Effects of Phosphorus

1.1.1 Too much phosphate can cause health problems, such as kidney damage and osteoporosis.

1.2 Environmental Effects of Phosphorus

1.2.1 In soil, phosphorus will remain for several days, but in deep soils and the bottom of rivers and lakes, phosphorus will remain for a thousand years or so. Orthophosphates applied to agricultural or residential cultivated land as fertilizers are carried into surface waters with storm runoff.

1.2.2 The increasing phosphorus concentrations in surface water, raises the growth of phosphate-dependent organisms, such as algae and duckweed. These organisms use great amounts of oxygen and prevent sunlight from entering the water. This is known as eutrophication.

2.0 Working Range: 0.05 – 1.0 mg/L

3.0 Summary

Ammonium molybdate and antimony potassium tartrate reacts in an acid medium with dilute solutions of phosphorus to form an antimony-phospho-molybdate complex. This complex is reduced to an intensely blue-colored complex by ascorbic acid. The color is proportional to the phosphorus concentration. Only orthophosphate forms a blue color in this test. Organic phosphorus compounds may be converted to the orthophosphate form by persulfate digestion.

4.0 Sample Preservation

4.1 The samples should be preserved by adding 0.5 mL/L of 1:1 H_2SO_4 as specified in CFR part 136 Table II. Samples may be stored in the refrigerator for up to 28 days.

4.2 Samples that are received preserved are checked for proper pH by the analyst prior to analysis.

1.1.1 Using a glass rod, place a small amount of sample on a pH strip

1.1.2 If the sample is at the proper pH place a **checkmark** ✓ in the pH column on the prep sheet.

1.1.3 If more preservative is required add sulfuric acid. Record ++ in the pH column and the qualifier (N_pAdl) must be attached to the sample result.

Note: Samples that are preserved by the splitter do not require analyst pH verification. Instead the analyst places a **dash –** on the prep sheet in the pH column.

5.0 Interferences

5.1 Copper, iron, or silicate at concentrations many times greater than their reported concentration in seawater causes no interference. However, high iron concentrations can cause precipitation of and subsequent loss of phosphorus.

5.2 Arsenate is determined similarly to phosphorus reacting with molybdate reagent to produce a blue color and should be considered when present in concentrations higher than phosphorus. Concentrations as low as 0.1mg As/L can interfere.

5.3 Hexavalent chromium and nitrite interfere to give results about 3% lower at concentrations of 1 mg/L and 10-15% lower at 10 mg/L.

6.0 Safety

Safety glasses and gloves must be worn when dealing with acids and bases.
See SOP S01 – Concentrated Acids and Bases
SOP S03 – Spill Control Policy

7.0 Equipment

7.1 Autoclave: Log# 225 - Autoclave #4, Log#8 – Autoclave #2, and Log#132 – Autoclave #3
7.2 100mL beakers
7.3 60mL culture tubes and silver caps.

7.4 Spectrophotometer: Spectronic Genesys 10 uv -1 (Log #152) and Genesys 10 uv -2 (Log#227)
7.5 Syringes and a .45 micron filter disk

8.0 Glassware Preparation

8.1 Glassware is stored separately for Total Phosphorus use only.

8.2 All glassware is acid washed in 1:1 HCl immediately after use, followed by thorough rinsing with three portions of D.I. water. If glassware still appears dirty, further steps are taken, by use of one of the following: Alconox and hot water, 1:1 acid rinse, acetone or appropriate solvent rinse. Glassware is always finished with a final D.I. rinse.

9.0 Phosphate Reagents and Standards

9.1 Reagent water: Nanopure

9.2 Potassium Persulfate, $K_2S_2O_8$: Kept at room temperature for up to 10 years.

9.3 P-Reagent

9.3.1 Ascorbic Acid solution: Dissolve 0.53 g Ascorbic Acid in 30 mL Nanopure water. Make fresh each time.

9.3.2 5N Sulfuric Acid: Slowly with stirring, add 70 mL concentrated H_2SO_4 to 430 mL nanopure water. Let cool and dilute to 500 mL. Reagent is stored in a glass bottle at room temperature for up to one year.

9.3.3 Ammonium Molybdate solution: Dissolve 20 g of Ammonium Molybdate 4-hydrate $[(NH_4)_6Mo_7O_{24}\cdot 4H_2O]$ in 500 mL Nanopure water. Reagent is stored in a glass bottle at room temperature for up to three months.

9.3.4 Potassium Antimonyl Tartrate: Dissolve 1.3715 g $K(SbO)C_4H_4O_6\cdot 1/2H_2O$ in 400 mL Nanopure water. Dilute to 500 mL Nanopure water. Reagent is stored in a glass bottle at room temperature for up to six months.

9.3.5 Mix in a beaker 50 mL 5N Sulfuric Acid, 5 mL Potassium Antimonyl Tartrate solution, and 15 mL of the Ammonium Molybdate solution. To this, add 30 mL Ascorbic Acid solution. This makes 100 mL P Reagent.

9.3.6 Let all reagents reach room temperature before mixing. Mix in the order given. If turbidity forms in the combined reagent, shake and let stand for a few minutes until turbidity disappears before proceeding. The reagent is stable for 4 hours at room temperature.

If refrigerated, the reagent can be kept and used for 3 days assuming all QC samples meet the proper acceptance criteria. Usually this reagent is kept no longer than 2 days. Place reagent immediately back in the refrigerator after use to preserve its integrity. When fresh the reagent should be light yellow. If the reagent turns gold, it is too old. Reagent is stored in glass.

9.3 Phenolphthalein Indicator: Dissolve 1 g of Phenolphthalein in 100 mL of reagent alcohol: ethyl or isopropyl: kept at room temperature for up to 1 year. Add 100 mL nanopure water. Indicator is stored in a glass bottle at room temperature for up to six months.

9.4 6N Sodium Hydroxide: Dissolve 240 g NaOH into 900 mL nanopure water. Stir on magnetic stir plate under fume hood. (Caution: sample will become hot, fumes are noxious.) Let cool. Dilute to 1 liter. Reagent is stored in a Nalgene bottle at room temperature for up to one year.

9.5 Sulfuric Acid Solution (30%): Gradually, add 300 mL concentrate sulfuric acid to 1000 mL nanopure water, cool. Warning, solution will get HOT. Reagent is stored in a glass bottle at room temperature for up to one year.

Note: The curve standards, Laboratory control samples, and blanks are all made with Nanopure water.

9.6 Laboratory Control(0.5 ppm):

9.6.1 Stock: 100 ppm solution containing organic phosphate, purchased from a certified vendor. Solution is stored at room temperature until manufacturer specified expiration date. Once opened the expiration date is 1 year from date opened as long as this does not exceed original manufacturer expiration date.

9.6.2 Working LCS (0.5 ppm): Pipette 50 uL of 100 ppm stock standard into 10 mL Nanopure water. Digest 10 mL of working standard to yield a 0.5 ppm LCS.

9.6.3 Calibration Check (0.25 ppm): Pipette 25 uL of 100 ppm Stock standard into 10 mL Nanopure water and digest in the autoclave.

9.6.4 MS/MSD (0.5 ppm): Pipette 50 uL of the 100 ppm Stock Standard into 10 mL of a sample, or to a dilution of sample with a final volume of 10 mL and digest in the autoclave.

9.7 Calibration Standards for Curve:

9.7.1 Stock: KH_2PO_4 previously dried in a 105^{o}C oven and stored in a desiccator.

9.7.2 Intermediate 1000 ppm: Weigh up 4.394 g KH_2PO_4 into 1 L of Nanopure. Keep at room temperature. This standard may be kept for up to 12 months or replaced sooner if comparisons with QC samples indicate a problem.

9.7.3 Working Standards:

9.7.3.1 Pipette 1 mL of 1000 ppm PO4-P Intermediate standard into 1 L of nanopure water to yield a concentration of 1ppm.

9.7.3.2 Once a year or when calibration checks indicate a need, digest various amounts of above solution to create a six-point curve. Solutions are made fresh each time.

9.7.3.3 The 1ppm Intermediate standard is diluted to the following concentrations: 0.05 ppm, 0.1 ppm, 0.3 ppm, 0.5 ppm, 0.75 ppm, and 1.0 ppm. See curve data for more information.

10.0 Procedure for Total Phosphorus:

10.1 Digestion

10.1.1 Transfer 10 mL of well-mixed sample (preserved with Sulfuric acid) into each culture tube.

10.1.2 Transfer 10 mL of nanopure to the culture tube for the Method Blank, Lab Control Spike, and CCV.

10.1.3 Spike 50 ul of 100 ppm LCS stock standard into 10 mL nanopure water for the Lab Control, Matrix Spike, and Matrix Spike Duplicate. (See section 9.6).

10.1.4 Spike 25 ul of 100 ppm LCS stock standard into 10mL nanopure water to yield a concentration of 0.25 ppm. (See section 9.6.3).

10.1.5 Add 200 ul of 30% Sulfuric Acid Solution (see section 9.5) to all culture tubes.

10.1.6 Add a scoop (0.05 g) of $K_2S_2O_8$, Potassium Persulfate, to all culture tubes.

10.1.7 Cover the culture tubes with silver caps and place in the autoclave in the Microbiology lab. Check that water is filled to the appropriate level. Document which autoclave was used on your data sheet.

10.1.8 Set the autoclave timer for 30 minutes slow mode (with cool time it is about 55 minutes. Heat at 98 to137 kPa. Autoclave reaches 15 psi which is equal to about 103 kPa.

10.1.9 Once cooled, take out of autoclave and pour sample from culture tubes to acid rinsed beakers. Rinse the culture tubes 3 times with nanopure slightly, not too much.

10.1.10 Add a drop of phenolphthalein indicator to each beaker.

10.1.11 Neutralize with 6N Sodium Hydroxide by adding one drop at a time using a plastic dropper until it turns pink.

10.1.12 Bring up to 20 mL with nanopure in a graduated cylinder and then pour back to the acid rinsed beaker. Check for tubidity at this time. Autoclave again with less volume if turbid.

10.2 Colorimetric Reading

10.2.1 Add 3.2 mL P-reagent, swirl, and let stand for minimum of 10 minutes. Read absorbance after 10 minutes but no more than 30 minutes.

10.2.2 Set Spectrophotometer wavelength to 880 nm.

10.2.3 Set zero with digested blank.

10.2.4 Read ABS of standard and record.

10.2.5 Record absorbance of samples. Rinse cuvette with nanopure in between samples. Rinse cuvette with sample prior to reading. If result is over 1 mg/L, dilute, and re-digest a new aliquot of sample.

10.2.6 Make sure to clean/dry off cuvette with tissue.

11.0 Calibration:

11.1 A six-point curve plus the zero point, required by the method, is analyzed on a yearly basis covering the referenced concentration range. (See 9.7.3 for more information).

11.2 An acceptable curve yields an $r^2 = 0.99$ or better.

11.3 A regression equation is generated from the curve.

11.4 Results are calculated from the regression equation.

11.5 Immediately following calibration a 0.5 mg/L ICV is analyzed using the LCS source to verify the curve. Results must be within 85-115% of the expected value or a new curve may be necessary.

11.6 Continuing Calibration Verification Checks: The calibration is verified on a daily basis by analysis of the Lab Control and a 0.25 mg/L Calibration Check analyzed at the end of the day's batch. Recovery of these checks must be within *85-115*% of expected value.

11.7 A standard at the reporting limit is analyzed at the end of the run. This is used to validate samples reported as none detected and to evaluate instrument sensitivity should calibration checks show a low bias. This standard should have a signal greater than the method blank.

12.0 Quality Control

Note: See also ESB SOP Q01 for general QC requirements

12.1 Laboratory Control (Conc 0.5 ppm):

12.1.1 A LCS is analyzed once per batch, or for every 20 samples, whichever is greater and reported per matrix type.

12.1.2 The results must be within the acceptance range of *85-115%* or the analysis is considered to be out of control. The problem must be corrected before the analysis can continue.

12.2 Matrix Spike and Matrix Spike Duplicate (Conc 0.5ppm):

12.2.1 A MS/MSD is analyzed once per batch, or 1 for every 20 samples per matrix type, whichever is greater.

12.2.2 If the matrix spike and matrix spike duplicate are not within the control limits of 80-120% with a maximum RPD of 20%, the spikes should be re-analyzed.

12.3 A Reagent Blank is digested with each batch of 20 samples and used to zero the instrument.

12.4 An MDL study is completed whenever major equipment or procedural changes are made. Standards are spiked at the reporting limit or 2.5-5 times the estimated MDL and a minimum of seven replicates is analyzed. See QA Manual for calculation. Results must be less than the reporting limit.

12.4.1 LOD Verification: On a yearly basis, a vial of reagent water is spiked at a level of at least 2-3 times analyte MDL. The sample is analyzed. Analyte response must be greater than method blank response to verify the Level of Detection or MDL.

12.5 Demonstration of Capability: Prior to initial analysis of samples or when a significant change is made to the method, a Demonstration of Capability Study is performed. This is accomplished by analysis of four replicates of

a QC sample made at a concentration 10 times the MDL to midrange of the curve. The average percent recovery of the QC samples must be 80-120% with a maximum %RSD of 20.

12.6 Demonstration of Continuing Proficiency: On an annual basis, analyst must turn in valid LCS data from four consecutive LCS samples or results from a successful Performance Evaluation Study. LCS percent recovery must meet laboratory prescribed acceptance criteria. Relative standard deviation between the four replicates must be less than or equal to 20%. Performance Evaluation Studies are performed twice a year.

13.0 Calculations

13.1 For Total Phosphorus

13.1.1 Convert Absorbance to concentration by plugging the spectrophotometer reading into a regression equation. Do not report results higher than the highest calibrator.

13.1.2 Enter initial digestion volume used for samples on the Bench Sheet.

13.1.3 Prepared date and time is documented as the date and time that the digestion reagents are added to the last sample of the batch.

13.1.4 Analysis date and time is documented as the date and time that the last sample absorbance was read.

13.1.5 Enter raw result into the LIMS.

13.1.6 Results are reported in mg/L to 2 significant figures.

13.1.7 Results below 0.05 mg/L are reported as ND. See LIMS for MDL information.

13.2 Other Forms of Phosphorus

13.2.1 Total Dissolved Phosphorus (TPD) – Sample is filtered thru a 0.45 micron filter prior to analysis. *To test the filters include a blank and BS with the analysis that has also been filtered.*

13.2.2 Total Insoluble Phosphorus (TPI) – Total Phosphorus minus Total Dissolved Phosphorus.

13.2.3 Organic Phosphorus (ORGP) = Total Phosphorus minus phosphorus generated from digest SM 4500-P B 2 below.

13.2.4 Acid Hydrolyzable Phosphorus (PAHYDRO) - see Standard Methods 4500-P B 2. Phosphorus resulting from this digest minus ortho phosphate = acid hydrolysable phosphorus.

13.2.5 Total Phosphate (PO4) = Total Phosphorus x3.

14.0 Definitions: See SOP Q15 – SOP Definitions

15.0 Corrective Action For Out of Control Or Unacceptable Data:

15.1 See SOP Q06 – Corrective Action

16.0 Pollution Prevention and Waste Management:

16.1 See SOP S05 – Neutralization Procedure for Acid and Alkaline Waste

16.2 SOP S06 – Disposal of Chlorinated Solvents

16.3 SOP S07 – Pollution Prevention

16.4 Wastes are dumped in the "Low Acid Concentration Waste" barrel or alternatively sample waste is placed in a stoppered sink, neutralized and then washed down the drain.

17.0 Method Performance

17.1 Refer to MDL studies, Initial Demonstration of Capability Studies, and laboratory control charts maintained in the QA Office.

References

Standard Methods for the Examination of Water and Wastewater, AWWA/APHA/WEF, 22nd Edition 4500-P 5B, E

GENESYS 10 Operator's manual Thermo Electron Corporation

Methods of Chemical Analysis of Water and Wastes, EPA Method 365.2

Note: All *italicized items* are an indication of a variation from the method.

Approved by: Julia Sudds Date: 04/03/14

AUTOMATED KJELDAHL NITROGEN
Edward S. Babcock & Sons
STANDARD OPERATING PROCEDURE
EPA Method 351.2
Effective Date: **__8/18/09_____**

1.0 Scope and Application:

1.1 This method covers the determination of total Kjeldahl nitrogen in drinking and surface waters, domestic and industrial wastes. This method may also be used for solid samples if both client and regulator agree to this variation.

1.2 Analytically organic nitrogen and ammonia are determined together as Kjeldahl nitrogen. Organic nitrogen includes such natural materials as proteins and peptides, nucleic acids and urea, and numerous synthetic organic materials. Typical organic nitrogen concentrations vary from a few hundred ug/L in some lakes to more than 20 mg/L in raw sewage. This procedure converts nitrogen components of biological origin, but may not convert all the nitrogenous compounds of some industrial wastes.

1.3 The organic nitrogen concentration is actually the total Kjeldahl nitrogen concentration minus the ammonia concentration. Organic nitrogen may be either dissolved or suspended particulate matter in water. High levels or organic nitrogen in water may indicate excessive production or organic pollution from the watershed. Animal and human waste, decaying organic matter and live organic material like tiny algae cells can cause organic nitrogen enrichment of lake water.

2.0 Working Range:

0.1mg/L to 3.0mg/L. This range is for photometric measurements made at 660nm in a 10 mm tubular flow cell. Higher concentrations can be determined by sample dilution.

Matrix	Reporting Limit	Method Detection Limit
Water/liquid	0.1mg/L	0.063mg/L
Solid/Sludge	100mg/kg	100mg/kg

3.0 Summary:

The sample is heated in a block digester in the presence of sulfuric acid, potassium sulfate and copper sulfate. The residue is cooled and diluted with Nanopure water. The ammonium ion is determined using an automated colorimetric analyzer.

4.0 Collection, Preservation, and Holding Time:

4.1 Samples are preserved to pH<2 with sulfuric acid. They are stored at 4 degrees Celsius.

4.2 Samples must be analyzed within 28 days.

4.3 Samples that are received preserved are checked for proper pH by the analyst prior to analysis.

4.3.1 Pour a small amount of sample over a pH strip

4.3.2 If the sample is at the proper pH place a **checkmark** ✓ in the pH column on the prep sheet.

4.3.3 If more preservative is required add sulfuric acid. Record ++ in the pH column and the qualifier (N_pAdl) must be attached to the sample result.

Note: Samples that are preserved by the splitter do not require analyst pH verification. Instead the analyst places a **dash –** on the prep sheet in the pH column.

5.0 Interferences:

5.1 The acid and salt content of the digestion reagent is intended to produce a digestion temperature of about 360 degrees Celsius. If the sample contains a very large quantity of salt or inorganic solids that dissolve during digestion, the temperature may rise causing pyrolytic loss of nitrogen. To prevent this from occurring, the analyst may use less sample.

5.2 If large amounts of organic matter are present an increase in digestion temperature may also occur. To prevent this from occurring, the analyst may use less sample volume.

5.3 Be cautious of turbidity that may result in the SEAL tubes from the boiling stones if the digestate is not properly decanted. Once the digested KJN samples have been poured into the SEAL tubes, discard the remaining digestate to prevent such problems.

5.4 The color reaction chemistry for TKN analysis is sensitive to changes in acid concentration in the diluted digests. The acid concentration may vary from the nominal value for various reasons. Typically-observed symptoms are significant change in the calibration absorbance range or excessive upward or downward curvature of the calibration line.

5.5 High nitrate concentrations (10x or more than the TKN level) result in low TKN values. If interference is suspected, samples should be diluted and reanalyzed.

6.0 Safety:

6.1 All digests must be performed under the hood. Take care when removing digests from the Block digester. Use gloves and safety goggles.

6.2 The following chemicals have the potential to be highly toxic or hazardous, consult the MSDS for more information that is located in the filing cabinet in the study.

6.2.1 Sulfuric Acid (Sections 8.7)

6.2.2 Sodium Nitroprusside (Sections 8.13 and 8.14)

6.3 See SOP S01 – Concentrated Acids and Bases
SOP S03 – Spill Control Policy

7.0 Apparatus:

7.1 Standard laboratory glassware: volumetric flasks, beakers-tall 200mL capacity, graduated cylinders, pipettes.

Note: Prior to use, glassware is acid rinsed with 1:1 HCl and three rinses of D.I. All glassware is cleaned immediately after use by thorough rinsing with three portions of D.I. water. If glassware still appears dirty, further steps are taken, by use of one of the following: Alconox and hot water, 1:1 acid rinse, acetone or appropriate solvent rinse. Glassware is always finished with a final D.I. rinse.

7.2 Block Digester: SEAL BD-46

7.3 SEAL – Discrete Automated Colorimetry

7.4 Vortex

7.5 Hengar Granules

8.0 Standards and Reagents:

8.1 Reagent water: Prepare all intermediate standards and reagents (including dilutions) in ammonium free (D.I.) water.

8.2 Laboratory Control Sample (LCS):

8.2.1 Stock LCS: 100 mg/L Stock TKN Standard purchased from ERA. Solution is stored at room temperature. Manufacturer expiration dates are observed. Once a stock standard is opened, the expiration date is one year as long as this date does not exceed the original manufacturer's date. Stock standards can be replaced sooner if comparisons with QC samples indicate a problem.

8.2.2 LCS: Digest 0.2mL Stock LCS. Dilute to 20mL with Nanopure water after digestion for a final concentration of 1mg/L.

8.2.3 Acceptability: If the LCS doesn't fall within the acceptance range of 80% to 120% recovery, the analysis is stopped until the cause is determined and the LCS is in control.

8.3 Matrix Spikes:

8.3.1 Spiking solution: Pipette 0.2mL of stock LCS on top of sample prior to digestion for MS. The spike concentration is 1 mg/L ammonia nitrogen in solution.

8.4 Calibration Standard:

8.4.1 Stock: Ammonium chloride (NH_4Cl)

8.4.2 Intermediate Standard (1000ppm):

8.4.2.1 Dehydrate Ammonium chloride (NH_4Cl) in a 105°C oven. Allow to cool in a dessicator.

8.4.2.2 Weigh out 3.819 g Ammonium Chloride. Dilute to 1 liter with D.I. water in a volumetric flask.

8.4.2.3 Pour the solution into a 1 liter amber bottle. Keep out of sunlight. Solution is stored at room temperature for up to one year. Standards can be replaced sooner if comparisons with QC samples indicate a problem.

8.4.3 Intermediate Standard A (50ppm): Dilute 0.5mL of above intermediate standard (8.4.2) up to 10mL with digested digestion reagent. This solution is prepared every 2 weeks and stored at room temperature.

8.4.4 Working Calibration Standards: Fresh standard is made every 2 weeks and stored at 4°C. *A 3ppm standard is diluted by adding 600 uL of 50ppm Intermediate standard up to 10 mL with digested digestion reagent*. The other *standards are diluted from the 3ppm standard by the instrument with digested digestion reagent.* Working Calibration standards concentrations are: 3.0, 2.0, 0.99, 0.51, 0.24 and 0.09 mg/L.

8.5 ICV and CCV: A second stock source of NH_4Cl is purchased. An intermediate standard (1000ppm) is prepared as specified in section 8.4.2. Standards are prepared as follows:

8.5.1 Standard A (50ppm): Digest 0.5mL of above intermediate standard and dilute to 10mL with digested digestion reagent. Fresh standard is made every 2 weeks and stored at room temperature.

8.5.2 Working Standards: Solutions are stored at 4°C for up to 2 weeks.

8.5.2.1 CCV - 2.0 mg/L standard: 400uL of 50ppm Intermediate into 10mL of digested digestion reagent.

8.5.2.2 ICV - 1.0 mg/L standard: 200 uL of 50ppm Intermediate into 10mL of digested digestion reagent.

8.5.2.3 RL Check - 0.1 mg/L standard: 0.5 mL of 2ppm CCV into 10mL of digested digestion reagent.

Note: All salts used to make the following reagents are stored at room temperature for up to 10 years from date received.

8.6 Stock sodium hydroxide (10% w/v)

8.6.1 Sodium hydroxide (NaOH) - 50 g

8.6.2 Dilute to 500 mL with DI water.

8.6.3 Dissolve 50 g sodium hydroxide pellets in 400 mL ammonia-free deionized water. Caution, the flask will become warm! Cool and dilute to 500 mL.

8.6.4 Store at room temperature for up to six months.

8.7 Digestion reagent
- 8.7.1 Potassium sulfate (K2SO4) - 134 g
- 8.7.2 Concentrated sulfuric acid (H2SO4) - 134 mL
- 8.7.3 Copper sulfate ($CuSO_4 \cdot 5H_2O$) - 11.4g
- 8.7.4 Dilute to 1 L with DI water.
- 8.7.5 Add 134 g potassium sulfate (K2SO4) to about 700 mL of deionized water and carefully add 134 mL concentrated sulfuric acid (H2SO4). Caution, the container will become hot! Add 11.4g $CuSO_4 \cdot 5H_2O$ and dilute to 1 Liter.
- 8.7.6 Store at room temperature for up to six months.

8.8 Stock sodium potassium tartrate solution (10% w/v)
- 8.8.1 Sodium potassium tartrate - 50 g
- 8.8.2 Dilute to 500 mL with DI water.
- 8.8.3 To prepare this reagent, dissolve 50 g sodium potassium tartrate in 400 mL ammonia-free deionized water and dilute to about 500 mL. Add 3 to 5 pellets of sodium hydroxide. Boil the solution, with stirring, for at least 1 hour to drive off ammonia contamination. Add make-up water if needed. Cover the flask and cool to room temperature. Add acid dropwise (sulfuric or hydrochloric, about 5 normal) to reach a pH of 7.5 ± 0.5. Dilute to 500 mL.
- 8.8.4 Store in the refrigerator up to six months.

8.9 Stock buffer solution
- 8.9.1 Sodium phosphate, dibasic anhydrous (Na2HPO4) -- 67 g
- 8.9.2 Sodium hydroxide - 10 g
- 8.9.3 Dilute to 500 mL with DI water.
- 8.9.4 Dissolve 67 g sodium phosphate, dibasic anhydrous (Na2HPO4) or 127 g sodium phosphate, dibasic heptahydrate (Na2HPO4•7H2O) in about 350 mL ammonia-free water. Add 10.0 g sodium hydroxide and stir to dissolve. Warm gently to dissolve and bring volume to 500 mL.
- 8.9.5 Store at room temperature for up to six months.

8.10 Working buffer solution
- 8.10.1 10% Sodium potassium tartrate solution (above) – 250 mL
- 8.10.2 Stock buffer solution (above) - 100 mL
- 8.10.3 10% Sodium hydroxide stock solution (above) – 120 mL
- 8.10.4 Dilute to 500 mL with DI water.
- 8.10.5 Combine reagents in the stated order; add 100 mL stock buffer solution to 250 mL 10% sodium potassium tartrate solution and mix. Add 10% sodium hydroxide stock solutionl. Dilute to 500 mL and invert to mix.
- 8.10.6 Store at room temperature for up to six months.

8.11 Alkaline sodium salicylate stock solution
- 8.11.1 Sodium salicylate, anhydrous - 75 g
- 8.11.2 Sodium hydroxide - 10 g
- 8.11.3 Dilute to 500 mL with DI water.
- 8.11.4 Add 10 g sodium hydroxide pellets to about 400 mL ammonia-free water and stir to dissolve. Add 75 g sodium salicylate and stir to dissolve. Dilute to 500 mL.
- 8.11.5 Store this solution in an opaque bottle. Store for up to 3 months or discard if the solution darkens significantly.

8.12 Sodium hypochlorite reagent (6.15% NaOCl (w/v)) Important: The bleach should not contain any additives other than NaOCl.
- 8.12.1 Refrigerate this reagent. Replace this solution every 4 months.
- 8.12.2 Dilute 10 mL, respectively, of purchased bleach to 50 mL with D.I.
- 8.12.3 Prepare fresh daily.

8.13 Stock sodium nitroprusside solution (30 g/L)
- 8.13.1 Sodium nitroprusside - 3 g
- 8.13.2 Dilute to 100 mL with DI water.
- 8.13.3 Dissolve 3 g sodium nitroprusside in 100 mL ammonia-free water.
- 8.13.4 Store this solution in an opaque bottle in the refrigerator. Replace monthly or sooner if a blue-green tint is seen.

8.14 Working salicylate/nitroprusside solution
- 8.14.1 Stock sodium nitroprusside solution (above) 0.25 mL
- 8.14.2 Stock sodium salicylate solution (above) 40 mL
- 8.14.3 Add 0.25 mL stock sodium nitroprusside solution and 40 mL stock sodium salicylate solution. Install the pierced cap and mix gently yet thoroughly over a sink.
- 8.14.4 Replace this reagent wedge every 3 days.

8.17 Concentrated Sulfuric Acid: Stored at room temperature for up to 10 years.

Note: The above reagent recipes are specified by the instrument manufacturer (SEAL AQ2 Method No: EPA-125-A Rev 3).

9.0 Procedure:

9.1 Digestion:

- 9.1.1 Add 3-4 Hengar Granules (use once only)
- 9.1.2 Pipet 5mL of digestion reagent into each tube.
- 9.1.3 Add 0.2mL of LCS spike to the LCS and Matrix spike samples.
- 9.1.4 Pipet 20mL of aqueous sample into each block digestion tube or use a dilution as needed. Do not add reagent water to tube even if sample is diluted. The method blank will contain digestion reagent only.
- 9.1.5 Make sure tubes are dry on the outside surfaces. Place digestion tubes into the block.
- 9.1.6 Flip the stand by switch to On. The yellow light will appear. It will take about 20-30 minutes to reach 180^{o}C. When the red light goes on indicating it has reached temperature, press the green Start button. The green light will go on. Samples will heat for about one hour evaporating the water.
- 9.1.7 When the buzzer sounds, press the green start button again and add the cold fingers.
- 9.1.8 After 1½ hours at 380^{o}, the buzzer will sound and the block will cool down to 180^{o}C.
- 9.1.9 Remove fingers and digestion tubes.

9.1.10 After a few minutes, while samples are still warm, add 19.3mL of Nanopure with a graduated cylinder to each sample and spin with vortex to mix.
9.1.11 Allow at least 2 hours for the turbidity to settle.
9.1.12 Pour into cuvettes, cool, and analyze on the SEAL.
9.1.13 See pages 17 – 20 in BD-46 User Manual for more details of Block Digestion procedure.

9.2 Solid Samples:

9.2.1 Weigh up 0.1g of solid sample into digestion vessel.
9.2.2 LCS 1ppm: 800 uL of 100ppm LCS Stock (section 8.2.1)
9.2.3 Sample duplicate at a frequency of 10% or per batch whichever is greater. Acceptance criteria: RPD maximum 25%.
9.2.4 Add 20mL of digestion reagent.
9.2.5 Continue digest starting in section 9.1.4.
9.2.6 After digestion, while samples are still warm, bring up to 80mL with Nanopure and spin with vortex to mix.
9.2.7 Pour into cuvettes, cool, and analyze on the SEAL.

9.3 Leaf Tissue (N):

9.3.1 Wash leaves with D.I. water.
9.3.2 Dry in feed oven overnight.
9.3.3 Grind leaves into a powder.
9.3.4 Digest sample according to section 9.2.
9.3.5 Report result as %N.

9.4 Analysis:

9.4.1 See SEAL SOP I41 for general operating instructions.
9.4.2 Load standard, reagents and samples.
9.4.3 Choose method: TKN.
9.4.4 Type in sample table.

9.5 Calibration

9.5.1 Calibrate the instrument every run or whenever the calibration check indicates a need. A 3 point curve including zero is required by method. Normally the laboratory performs a 6-point curve, plus a blank. An acceptable curve yields an r = 0.995 or better. To obtain the best possible curve fit, the zero point is forced, included, or ignored depending on analyte performance.
9.5.2 The Linear Calibration Range is proven *with each calibration* since 6 points are used and the curve is linear. Results are not reported over the highest calibrator, see 11.3. Immediately following calibration, an initial calibration verification standard from a second source (can be the LCS) is analyzed to verify the calibration. It must be within 90-110% recovery limits or a new calibration will be performed.
9.5.3 Calibration Check Standards: Prepare standards at 1.0ppm and 2.0ppm (section 8.5) to check the calibration. Analyze prior to sample analysis every 10 samples and at the end of the run to

prove that the existing calibration is still valid. Results must be within *85 – 115% recovery*.

9.5.4 A calibration Blank is analyzed after calibration, every 10 samples and at the end. Results must be less than the reporting limit.

9.5.5 A standard at the reporting limit is analyzed at the end of the run. This is used to evaluate instrument sensitivity should calibration checks show a low bias and to validate samples reported as none detected. This standard should have a signal greater than the method blank.

10.0 Quality Control:

Note: See also ESB SOP Q01 for general QC requirements

10.1 Matrix Spikes -- Matrix Spikes are performed on 10% of the samples in every analytical batch with a minimum of one MS performed per batch. . MS recovery will be compared to acceptance ranges *generated from historical data*. See LIMS for most current limits. If they do not fall within the acceptance range, the sample must be re-analyzed. If they still do not fall into the acceptance range, the data is flagged.

10.2 Laboratory Control -- A standard from a different source than the calibration standards shall be processed as a laboratory control with each analytical batch. One LCS will be run with every analytical batch and at a minimum of once for every 20 samples, per matrix type. This control will be compared to acceptance ranges of *80-120% recovery*. If the control does not fall within acceptance ranges, the analysis is considered to be out of control. No further samples shall be analyzed until the out of control condition is corrected.

10.3 Method Blank – A sample consisting of 5mL of digestion reagent is digested and analyzed as the method blank. One Method Blank will be run with every analytical batch and at a minimum of once for every 20 samples, per matrix type. It must read below but not more negative than the *RL* for batch acceptance. Samples results reported must be accompanied with a note if the method blank exceeds a concentration greater than 1/10 of the measured raw concentration of the sample. However blank results below the *RL* are considered to be ND and will not require a note.

Note: The blank value noted in the NBLK qualifier is the raw blank result. If the sample being qualified was diluted, in addition to NBLK, the analyst must also attach the qualifier NBLKd which includes the sample raw result.

10.4 An MDL study is completed whenever major equipment or procedural changes are made. Standards are spiked at the reporting limit or 2-3 times the estimated IDL and a minimum of seven replicates is analyzed. See QA Manual for calculation. Results must be below the reporting limit.

10.4.1 LOD Verification: On a yearly basis, a QC sample is spiked at a level of not more than 2-3 times analyte MDL. The sample is

analyzed. Analyte response must be greater than method blank response to verify the Level of Detection or MDL

10.5 Initial Demonstration of Capability: Prior to analysis of samples or when a significant change is made to the method, an Initial Demonstration of Capability Study is performed. This is accomplished by analysis of four replicates of a QC sample made at a concentration 1-4 times the RL. The average percent recovery of the QC samples must be 90-110% with a maximum %RSD of 20.

10.6 Demonstration of Continuing Proficiency: On an annual basis, analyst must turn in valid LCS data from four consecutive LCS samples or results from a successful Performance Evaluation Study. LCS percent recovery must meet laboratory prescribed acceptance criteria. Relative standard deviation between the four replicates must be less than or equal to 20%.

10.7 Performance Evaluation Studies are performed *twice* a year. A QCS sample acquired from an independent source such as ERA is analyzed. *Acceptance criteria supplied by the manufacturer is used to access performance.*

11.0 Calculations:

11.1 The instrument compares peak *areas* of samples, rather than peak height, against the calibration curve. Raw results are transferred into LIMS.

11.1.1 Digestion volumes are entered in LIMS as the initial and final volumes.
11.1.2 Dilutions of the digest are entered into the DIL column of LIMS.

11.2 If diluted samples read below 0.1 mg/L, re-analyze using more sample and diluting to a final volume of 20mL. Ideally diluted samples should fall in the middle or upper part of the calibration curve.

11.3 Do not report results higher than the highest calibrator. Digestate may be diluted in half with digested digestion reagent. If further sample dilution is necessary, re-digest a smaller aliquot.

11.4 If a result is more negative than the reporting limit, the digestate is diluted x2 and reanalyzed. If it is still too negative, the sample is redigested.

11.5 Watch for possible carry over. Studies have shown that carry over down to the MDL may occur over a concentration of 50mg/L. Rerun samples at or above the reporting limit following a sample at this concentration. Be mindful of carry over that may happen in the digestion block.

11.6 Organic Nitrogen (mg/L) = Kjeldahl nitrogen (mg/L) – Ammonia nitrogen (mg/L)

11.7 Total Nitrogen (mg/L) = Kjeldahl nitrogen (mg/L) + Nitrate nitrogen (mg/L) + Nitrite nitrogen (mg/L).

11.8 Report all results to two significant figures.

12.0 Definitions: See SOP Q15 – SOP Definitions

13.0 Pollution Prevention and Waste Management:

13.1Waste Disposal

13.1.1 Instrument waste is placed in the "KjN waste" barrel.

13.1.2 Sample waste after digestion is placed in the "Copper Waste" barrel.

13.2 See also SOP S05 – Neutralization Procedure for Acid and Alkaline Wastes
SOP S06 – Disposal of Chlorinated Solvents
SOP S07 – Pollution Prevention

14.0 Corrective Action For Out of Control or Unacceptable Data:

See SOP Q06 – Corrective Action

15.0 Method Performance

Refer to MDL studies, Initial Demonstration of Capability Studies, and laboratory control charts.

References:

Standard Methods for the Examination of Water and Wastewater APHA, AWWA, WPCF 18th Edition. 4500-O_{org}
Standard Methods for the Examination of Water and Wastewater APHA, AWWA, WPCF 20th Edition. 4500-O_{org}
SEAL AQ2 Instrument Manual and Method No: EPA-125-A Rev 3
EPA Method 351.2 (1993) Methods for the Chemical Analysis of Waters and Wastes.

Note: All *italicized items* are an indication of a variation from the method.

Approved by: Susann K. Thomas Date: 08/11/09

Standard Operating Procedure
Babcock Laboratories, Inc.
SM 4500-NH_3 G
Nitrogen, Ammonia (Colorimetric, Automated Phenate)
Effective Date: __04/07/14___

1.0 Scope and Application

1.1 This method covers the determination of ammonia in drinking, surface, and saline waters, domestic and industrial wastes in the range of 0.1 to 2.0 mg/L NH3 as N. This range is for photometric measurements made at 660 nm in a 10 mm tubular cell. Higher concentrations can be determined by sample dilution. This method may also be used for solid samples if both client and regulator agree to this variation.

1.2 Environmental Relevance: Ammonia is present naturally in surface and wastewaters. Its concentration is generally low in ground waters because it adsorbs to soil particles and clays and is not leached readily from soils. It is produced largely by deamination of organic nitrogen containing compounds and by hydrolysis of urea. At some treatment plants ammonia is added to react with chlorine to form combined chlorine residual. In the chlorination of wastewater effluents containing ammonia, virtually no free residual chlorine is obtained until ammonia has been oxidized. The chlorine reacts with ammonia to form mono and dichloramines.

1.3 Ammonia is the most reduced form of nitrogen and is found in water where dissolved oxygen is lacking. When dissolved oxygen is readily available, bacteria quickly oxidize ammonia to nitrate through nitrification. Other types of bacteria produce ammonia as they decompose dead plant and animal matter. Depending on temperature and pH, high levels of ammonia can be toxic to aquatic life. High ammonia concentrations can stimulate excessive aquatic production and indicate pollution. Important sources of ammonia to lakes and streams can include: fertilizers, human and animal wastes, and by-products from industrial manufacturing processes.

2.0 Summary of Method

2.1 Samples are distilled (if under NPDES program) or filtered and analyzed on an automated colorimetric system. Alkaline phenol and hypochlorite react with ammonia to form indophenol blue that is proportional to the ammonia concentration. The blue color formed is intensified with sodium nitroprusside.

3.0 Sample Handling and Preservation

3.1 Preservation by addition of conc. H_2SO_4 to a pH < 2 and refrigeration at 4°C.

3.2 Samples must be analyzed within 28 days.

4.0 Interferences

4.1 Calcium and magnesium ions, may be present in concentrations sufficient to cause precipitation problems during analysis. A 7.5% EDTA solution is used to prevent the precipitation of calcium and magnesium ions from river water and industrial waste. For sea water, a sodium potassium tartrate solution may be used. (See SM 4500-NH_3 H, 3e. for recipe.)

4.2 Sample turbidity and color may interfere with this method. Turbidity must be removed by filtration prior to analysis. Sample color that absorbs in the photometric range used will also interfere. If necessary, sample is diluted.

4.3 Urea and cyanates will hydrolyze on distillation at pH of 9.5.

4.4 Dechlorination is not necessary since the colorimetric analysis will detect chloramines formation.

4.5 Marked variation in acidity and alkalinity are eliminated by sample preservation with H_2SO_4. The pH is then checked to ensure that it is <2. Due to the reducing nature of this environment, residual chlorine is not expected to be a problem. *The sample is neutralized prior to analysis by the addition of the first reagent that is a NaOH buffer.*

4.5.1 For highly acidic samples the pH of the dilution water should be raised to approximately 11pH before adding the sample to avoid over diluting.

4.6 Distillation is required for all samples under the NPDES permit program. All other samples are filtered and may be distilled if the sample presents special matrix problems.

5.0 Apparatus

5.1 Test tube rack from SEAL.

5.2 13 x 100 mm disposable culture tubes.

5.3 SEAL Analyzer

5.4 Whatman 2 and Whatman 4 (11.0cm) filter paper or Gelmin 0.45 micron disk filters.

5.5 100 mL beakers.

5.6 1 mL, 5 mL, 10 mL autopipettes.

5.7 25 mL, 50 mL, and 100 mL graduated cylinders.

5.8 Distillation apparatus: #113 – Enviro Midi-Distiller Glastron, Inc.
#212 – Distiller Environmental Express.

5.9 VWR boiling chips 26397-409

5.10 Antifoaming Agent JT Baker B531-05

Note: Rinse all glassware prior to use with three portions of D.I. water. Immediately after use thoroughly rinse with three portions of D.I. water. If glassware still appears dirty, further steps are taken, by use of one of the following: Alconox and hot water, 1:1 HCl, acetone or appropriate solvent rinse. Glassware is always finished with a final D.I. rinse.

6.0 Reagents

6.1 Reagent Water: D.I. water

6.2 Sulfuric Acid solution: concentrated. Store at room temperature for up to 10 years.

6.3 Diluent or preserved water: Add 1 mL of Sulfuric acid to 1 L of D.I.. This solution is stored for up to 6 months at room temperature.

6.4 Sodium phenate: To a 100 mL beaker, add 2 g of sodium hydroxide and about 10 mL DI. Swirl to dissolve and cool to room temperature. In the fume hood, add 5 g of crystalline phenol and swirl to dissolve. Transfer to a 50 mL graduated cylinder and fill to the mark with DI water. Store in the refrigerator for three weeks or until the reagent becomes dark brown.

6.5 Sodium hypochlorite solution: bleach solution containing 6% NaOCl (such as "Clorox"). Store in refrigerator for up to 4 months.

6.6 Modified EDTA Buffer: Disodium ethylenediamine-tetraacetate (EDTA) (7.5%): Dissolve 18.75 g of EDTA (disodium salt) and 4.55 g of NaOH in 250 mL of D.I. water. This solution is stored for up to 6 months at room temperature.

6.7 Sodium nitroferricyanide (0.2%): Dissolve 0.5 g of sodium nitroferriocyanide in 250 mL of D.I. water. This solution is stored for up to 1 month at room temperature in a dark bottle. Discard if the reagent becomes bluish in color.

6.8 1 N NaOH: Dissolve 40 g of NaOH into 1 L of D.I. water. This solution is stored for up to 6 months at room temperature.

6.9 0.04N Sulfuric Acid: Add 1.12 mL of concentrated sulfuric acid into 1 L of D.I. water. This solution is stored for up to 6 months at room temperature.

6.10 Hellma Hellmanex II: This solution is stored at room temperature for up to 10 years.

6.10.1 Add 2 mL of Hellmanex solution into 100 mL of D.I. water. This solution is kept at room temperature for up to 3 months.

6.11 Borate Buffer:

6.11.1 0.1 N NaOH: Dissolve 4 g of NaOH into 1 L of D.I. water.
6.11.2 Mix 0.95 g $Na_2B_4O_7 \cdot 10H_2O$ into 100 mL of Nanopure water.
6.11.3 Add 8.8 mL of 0.1N NaOH and 50 mL of borate solution into a 100 mL flask and bring up to volume with D.I. water.
6.11.4 Solutions above are stored for up to 6 months at room temperature.

Note: The above reagent recipes are specified by the instrument manufacturer (SEAL Analytical AQ2 Method No: EPA 103-A Rev 4 © 6/01/05) and by Standard Methods 22nd Edition 4500-NH_3.

7.0 Standards and Quality Control Requirements:

Note: See also ESB SOP Q01 for general QC requirements

7.1 Lab Control Sample (LCS):

7.1.1 Stock Solution: 1000 mg/L NH_4 Standard - stored at room temperature using manufacturer specified holding times. Once a stock standard is opened, the expiration date is one year* as long as this date does not exceed the original manufacturer's date and percent recovery is acceptable.
(*one year may be extended if data is still acceptable)
7.1.2 LCS: Dilute 50 uL of stock standard (7.1.1) to 50 mL of diluent (6.3). The concentration is 1 mg/L NH_4 or 0.78 mg/L NH_4-N. (See below) This solution is stored at 4°C for up to 2 weeks.
7.1.3 An LCS is analyzed for every 20 samples per matrix type or one per analysis batch whichever is greater. LCSs are distilled unless the sample batch contains only undistilled samples.
7.1.4 If the LCS analysis does not fall within the acceptance range of 90-110%, the analysis is stopped until the cause is determined and the LCS is within the acceptance range.

$$1 \text{ mg/L } NH_4 \text{ X } \frac{14 \text{ g/mole N}}{18 \text{ g/mole } NH_4} = 0.78 \text{ mg/L N}$$

7.2 Matrix Spike (MS) / Matrix Spike Duplicate (MSD)

7.2.1 Spike solution: Spike 50 uL of stock standard (7.1.1) into 50 mL of sample. The spike concentration is 1 mg/L NH_4 or 0.78 mg/L NH_4-N.
7.2.2 A MS/MSD is analyzed for every twenty samples per matrix type or one per analysis batch whichever is greater. Spikes are distilled unless the sample batch contains only undistilled samples.
7.2.3 Acceptability: Acceptance ranges are 80-120% recovery with a maximum RPD of 20%.

7.3 Method Blank

7.3.1 Use diluent from section 6.3.
7.3.2 A MB is analyzed for every twenty samples per matrix type or one per analysis batch whichever is greater and reported per matrix type. Blanks are distilled unless the sample batch contains only undistilled samples.
7.3.3 Acceptability: MB must read below but not more negative than the RL of 0.1 mg/L for batch acceptance. Samples results reported must be accompanied with a note if the method blank exceeds a concentration greater than 1/10 of the measured raw concentration of the sample however, blank results below the RL are considered to be ND and will not require a note.

Note: The blank value noted in the NBLK qualifier is the raw blank result. If the sample being qualified was diluted, in addition to NBLK, the analyst must also attach the qualifier NBLKd which includes the sample raw result.

Note: Since the intensity of the color used to quantify the concentration is pH dependent, the acid concentration of the diluent and the standard ammonia solutions should approximate that of the samples.

7.4 Calibration Standard:

7.4.1 Stock: Ammonium chloride (NH_4Cl)
7.4.2 Intermediate Standard 1000 ppm:
7.4.2.1 Dehydrate Ammonium Chloride (NH_4Cl) in a 105°C oven.
7.4.2.2 Allow salt to cool in a dessicator. Weigh out 3.819 g NH4Cl.
7.4.2.3 Dilute to 1 liter with D.I. water in a volumetric flask containing 1.5 mL sulfuric acid.
7.4.2.4 Pour the solution into a 1 liter amber bottle. Keep at room temperature, out of sunlight. This standard may be kept for up to 1 year or replaced sooner if comparisons with QC samples indicate a problem.

7.4.3 Intermediate standard (50 ppm): Dilute 2.5 mL of 1000 ppm stock standard into 50 mL of diluent. This solution is stored at 4°C up to 28 days.

7.4.4 Dilute to make the following calibration standards:

7.4.4.1 2.0 mg/L standard: 2 mL of 50 ppm Intermediate into 50 mL of diluent.

7.4.4.2 The instrument dilutes the 2 ppm standard to the following concentrations: 0.02, 0.05, 0.1, 0.4, 0.8, and 1.6 ppm.

7.4.4.3 Calibration Blank: diluent.

Note: These solutions are stored at 4°C for up to two weeks.

7.5 ICV and CCV: A second stock source of NH_4Cl is purchased. An intermediate standard is prepared as specified in section 7.4.2 and section 7.4.3. Working standards are stored at 4°C for up to two weeks. Solutions are prepared as follows:

7.5.1 1.5 mg/L standard: 1.5 mL of 50 ppm Intermediate into 50 mL of diluent. Standard ran at the end of the run.

7.5.2 1.0 mg/L standard: 1 mL of 50 ppm Intermediate into 50 mL of diluent. Standard ran at the beginning, every 10 samples, and at the end of the run.

7.6 A calibration blank is analyzed at the beginning of the run, every 10 samples and at the end of the run. *Results must be less than but not more negative than the RL.*

7.7 A standard at the reporting limit is analyzed at the end of the run. This is used to validate samples reported as none detected and to evaluate instrument sensitivity should calibration checks show a low bias. This standard should have a signal greater than the method blank.

7.8 An MDL study is completed whenever major equipment or procedural changes are made. Standards are spiked at the reporting limit or 2.5-5 times the estimated MDL and a minimum of seven replicates is analyzed. See QA Manual for calculation. Results must be below the reporting limit.

7.8.1 On an annual basis perform the LOD verification check. Spike a QC sample of reagent water at a level of no more than 2-3 times analyte MDL. Analyte response must be greater than method blank response.

7.9 Demonstration of Capability: Prior to initial analysis of samples or when a significant change is made to the method, a Demonstration of Capability Study is performed. This is accomplished by analysis of four replicates of a QC sample made at a concentration 10 times the MDL to midrange of the curve. The average percent recovery of the QC samples must be 80-120% with a maximum %RSD of 20.

7.10 Demonstration of Continuing Proficiency: On an annual basis, analyst must turn in valid LCS data from four consecutive LCS samples or results from a successful Performance Evaluation Study. LCS percent recovery must meet laboratory prescribed acceptance criteria. Relative standard deviation

between the four replicates must be less than or equal to 20%. Performance Evaluation Studies are performed *twice* a year.

8.0 Procedure:

8.1 Sample Preparation

8.1.1 pH Verification

8.1.1.1 Samples that are received preserved are checked for proper pH by the analyst just prior to analysis.

8.1.1.1.1 Place a drop of sample onto a pH strip.

8.1.1.1.2 If the strip is red, the sample is pH<2. Place a **checkmark** ✓ in the pH column on the prep sheet.

8.1.1.1.3 If more preservative is required, add additional sulfuric acid and place a ++ on the prep sheet. Add the qualifier (N_pAdl) when the result is entered.

8.1.1.2 The analyst does not need to verify pH of samples that are preserved by the splitter. To indicate this, place a **dash –** on the prep sheet in lieu of a checkmark.

8.1.2 Distillation is required for all samples under the NPDES permit program. First check the distillation comparison study records. If a sample from the same site has been distilled in the past, and the RPD between the distilled and undistilled results is less than or equal to 20, then the sample does not need to be distilled. Raw results $\leq$1.0 mg/L will not generate useful RPD statistics. Instead these results must be within 0.1 mg/L of each other to eliminate the distillation step in the future. If a sample site is not in the study then it must be distilled and added to the study.

8.1.2.1 If the apparatus sits idle for an extended period of time or if samples have left a residue clean the apparatus as follows:

8.1.2.1.1 Rinse all glassware with D.I.

8.1.2.1.2 Make clean out solution by adding 25 mL of borate buffer to 500 mL of D.I. Adjust pH to 9.5 with 6N NaOH solution. Fine tune with 1N NaOH.

8.1.2.1.3 Fill distillation tubes to the line with clean out solution. Add boiling chips.

8.1.2.1.4 Distill over at least 30 mL

8.1.2.1.5 Re-rinse all glassware.

8.1.2.2 Then add 50 mL of sample, LCS, MS, MSD and diluent method blank to individual distillation tubes.

8.1.2.3 Add 2.5 mL of borate buffer to each flask.

8.1.2.4 Adjust to pH 9.5 with 1N or 6N NaOH.

8.1.2.5 Add several porous VWR boiling chips.

8.1.2.6 Distill at 190 ^{O}C into a receiving tube containing 5 mL of 0.04N sulfuric acid. Ensure that the tip of the delivery tube is placed below the sulfuric acid level.

8.1.2.7 Collect 50 mL of distillate. Note: Collect at least 30 mL then adjust to 50 mL with D.I. water.
8.1.2.8 Move the delivery tube out of the distillate; add extra water to the distillation tube to prevent dryness until the unit is turned off.
8.1.2.9 Turn off unit when the last sample is finished.
8.1.2.10 Pour distillate into SEAL tubes.

8.1.3 Undistilled samples
8.1.3.1 Filter samples if turbid through 0.45 micron microdisk filters directly into SEAL test tubes unless a dilution is made. If a dilution is required, dilute sample, bring up to 50 mL, and filter prior to pouring into SEAL tube. If less than 5 mL of sample is used, dilute with diluent otherwise D.I. water may be used.
8.1.3.2 Use the following volumes based on sample matrix:
8.1.3.2.1 Industrial or Influent Wastewater – 2-5 mL.
8.1.3.2.2 Effluent Wastewater – 25-50 mL.
8.1.3.2.3 Well water - 50 mL.
8.1.3.2.4 Solids – Make a 1:10 water extract, extract and swirl periodically for one hour. Filter prior to analysis.

8.2 Calibration:

8.2.1 The instrument is calibrated every run, when a major instrumentation change is made, or when the calibration check fails.
8.2.2 The analyst first analyzes a 1.0 mg/L standard. If the initial calibration check (ICV) is valid, within 90 – 110% of the expected value, a new calibration is not necessary.
8.2.3. If the ICV is out, calibrate the instrument. Although a 3-point curve is required, a 7-point curve plus blank is usually prepared. An acceptable curve yields an r = 0.995 or better.
8.2.4. Calibration Check Standard: Prepare a 1.0 ppm standard to check the calibration. Analyze prior to sample analysis, every 10 sample, and at the end of the run to prove that the existing calibration is still valid. A 1.5 ppm standard is analyzed at the end of the run. Recovery must be 90-110%.
8.2.5. Calibration Check Blank: Analyze a calibration blank prior to sample analysis, every 10 samples, and at the end of the run to prove that the existing calibration is still valid. *Results must be less but not more negative than the reporting limit.*
8.2.6. A standard at the reporting limit is analyzed at the end of the run. This is used to evaluate instrument sensitivity should calibration checks show a low bias. This standard should have a signal greater than the method blank

8.3 Analyze samples on the SEAL.

8.4 Allow instrument to warm up

8.5 Load standard, reagents and samples.

8.6 See SEAL SOP I41 for general operating instructions.

8.7 Choose method and begin analysis.

8.8 When an acceptable calibration has been performed, submit the tray of samples.

8.9 If diluted samples read below 0.1 mg/L, re-analyze using more sample and diluting to a final volume of 50 mL. Ideally, diluted samples should not fall in the lower quarter of the calibration curve.

8.10 If any sample reads above 2.0 mg/L, re-analyze using less sample.

8.11 Watch for possible carry over. Studies have shown that carry over may occur over a concentration of 50 mg/L. Rerun samples at or above the reporting limit following a sample at this concentration. Be mindful of carry over that may happen in the distillation apparatus.

9.0 Calculations

9.1 Enter raw results into the LIMS. Preparation aliquots are entered as the initial and final volumes. Dilution factors performed after preparation are entered in the DIL column in LIMS. The preparation time is set as the time the last sample in the batch was distilled or filtered. The analysis time is entered individually by the instrument during data transfer.

9.2 Aqueous reporting limit is 0.1 mg/L, Nonaqueous reporting limit is 1 mg/L. Refer to Element for MDL information.

9.3 Report 2 significant figures for samples and 3 significant figures for QC samples.

9.4 Inorganic Nitrogen = $NH_3N + NO_3N + NO_2N$

10.0 Definitions: See SOP Q15 – SOP Definitions

11.0 Safety: The toxicity or carcinogenicity of chemicals used in this method has not been precisely defined. Each chemical should be treated as a potential health hazard, and exposure to these chemicals should be minimized. A reference file of material data handling sheets is made available to all personnel involved in the chemical analysis.

11.1 The following chemicals have the potential to be highly toxic or hazardous, consult MSDS.
- 11.1.1 Sulfuric Acid
- 11.1.2 Phenol
- 11.1.3 Sodium nitroprusside

11.2 Safety Information about Enviro Midi-Dist Distillation System
- 11.2.1 Do not operate the Enviro Midi-Dist in the vicinity of combustible material.
- 11.2.2 During operation the surfaces around the heater assembly will get HOT- Do not touch the outer surface.
- 11.2.3 Do not move the Enviro Midi-Dist system while hot.
- 11.2.4 Do not attempt to operate the Enviro Midi-Dist system over 190 ^{O}C.
- 11.2.5 Allow the Enviro Midi-Dist to cool for 20minutes before removing glassware. Separation of the "hot" glassware components could result in bumping, boil over, and/or spraying of hot corrosive/caustic materials.
- 11.2.6 For more information see the Instruction Manual for the Enviro Midi-Dist Distillation System.

11.3 See SOP S01 – Concentrated Acids and Bases
SOP S03 – Spill Control Policy

12.0 Corrective Action For Out of Control or Unacceptable Data:
See SOP Q06 – Corrective Action

13.0 Pollution Prevention and Waste Management:
13.1Waste Disposal
- 13.1.1 Instrument waste is placed in the "KjN waste" barrel.
- 13.1.2 Sample waste after distillation is placed in the "High Acid Concentration Waste" barrel.
- 13.1.3 Unused distillate or filtrate is placed in the "Low Acid Concentration Waste" barrel.

13.2 See also SOP S05 – Neutralization Procedure for Acid and Alkaline Wastes
SOP S06 – Disposal of Chlorinated Solvents
SOP S07 – Pollution Prevention

14.0 Method Performance
Refer to MDL studies, Initial Demonstration of Capability Studies, and laboratory control charts maintained in the QA Office.

15.0 References

Standard Methods for the Examination of Water and Wastewater APHA, AWWA, WEF 22nd Edition 4500-NH_3 G

SEAL Analytical AQ2 Method No: EPA 103-A Rev 4 © 6/01/05

EPA Method 350.1. Methods for the Chemical Analysis of Waters and Wastes.

SEAL Analytical AQ2 Operations Manual

Note: All *italicized items* are an indication of a variation from the method.

Approved by: Julia Sudds Date: 04/03/14

METHOD #: 6010B
Edward S. Babcock & Sons Standard Operating Procedure
Effective Date: 3/31/14

TITLE: Inductively Coupled Plasma-Atomic Emission Spectroscopy

Table 1

ANALYTE:	CAS #	TTLC(TAD) ppm	STLC(WET) ppm	TCLP ppm
(Aluminum (Al))	7429-90-5			
Antimony (Sb)	7440-36-0	500	15	
Arsenic (As)	7440-38-2	500	5	5
Barium (Ba)	7440-39-3	10,000	100	100
Beryllium (Be)	7440-41-7	75	0.75	
(Boron (B))	7440-42-8			
Cadmium (Cd)	7440-43-9	100	1	1
(Calcium (Ca))	7440-70-2			
Chromium (Cr)	7440-47-3	2500	5	5
Cobalt (Co)	7440-48-4	8000	80	
Copper (Cu)	7440-50-8	2500	25	
(Iron (Fe))	7439-89-6			
Lead (Pb)	7439-92-1	1000	5	5
(Lithium (Li))	7439-93-2			
(Magnesium (Mg))	7439-95-4			
(Manganese (Mn))	7439-96-5			
(Mercury (Hg))	7439-97-6	20	0.2	0.2
Molybdenum (Mo)	7439-98-7	3500	350	
Nickel (Ni)	7440-02-0	2000	20	
(Potassium (K))	7440-09-7			
Selenium (Se)	7782-49-2	100	1	1
(Silica (Si))	7631-86-9			
Silver (Ag)	7440-22-4	500	5	5q
(Sodium (Na))	7440-23-5			
(Strontium (Sr))	7440-24-6			
Thallium (Tl)	7440-28-0	700	7	
(Tin (Sn))	7440-31-5			
Vanadium (V)	7440-62-2	2400	24	
Zinc (Zn)	7440-66-6	5000	250	

Analytes in parenthesis are not certified by NELAP.

INSTRUMENTATION: Inductively Coupled Plasma-Atomic Emission Spectroscopy
ICP-AES : Perkin Elmer Optima 5300DV

1.0 SCOPE AND APPLICATION

1.1 Inductively coupled plasma-atomic emission spectroscopy (ICP-AES) determines trace elements, including metals, in solution. The method is applicable to all of the elements listed in Table 1. All matrices, including ground water, aqueous samples, TCLP and CAM WET extracts, industrial

and organic wastes, soils, sludges, sediments, and other solid wastes, require digestion prior to analysis. Refer to the metals digestion SOP (M02) for more detailed information regarding digestion.

1.2 Table 1 lists the elements for which this method is applicable. Detection limits, sensitivity, and the optimum and linear concentration ranges of the elements can vary with the wavelength, spectrometer, matrix and operating conditions. Table 2 lists the recommended analytical wavelengths and estimated instrumental detection limits for the elements in clean aqueous sample matrices. The detection limit data may be used to estimate instrument and method performance for other sample matrices. Other wavelengths are used by our laboratory for some analytes. The detection limits are similar and they are relatively free of interferences (when compared with the recommended wavelengths).

1.3 The data quality objectives are stated and the initial demonstration of proficiency is documented and on file.

1.4 Use of this method is restricted to spectroscopists who are knowledgeable in the correction of spectral, chemical, and physical interferences described in this method.

1.5 Working range of analytes: Reporting limit to 90% of Linear Dynamic Range Maximum. See LIMS for most current RL and MDL.

2.0 SUMMARY OF METHOD

2.1 Prior to analysis, samples are solubilized or digested using appropriate Sample Preparation Methods (see SOP M02).

2.2 This method describes multielemental determinations by ICP-AES using sequential or simultaneous instruments. The instrument measures characteristic emission spectra by optical spectrometry. Samples are nebulized and the resulting aerosol is transported to the plasma torch. Element-specific emission spectra are produced by a radio-frequency inductively coupled plasma. The spectra are dispersed by a grating spectrometer, and the intensities of the emission lines are monitored by photosensitive devices such as diode arrays or photomultiplier tube(s). Background correction is utilized for trace element determination. Background must be measured adjacent to analyte lines on samples during analysis. The position selected for the background-intensity measurement, on either or both sides of the analytical line, will be determined by the complexity of the spectrum adjacent to the analyte line. The position chosen is as free as possible from spectral interference and should reflect the same change in background intensity as occurs at the analyte wavelength measured. Background correction is not required in cases of line broadening where a background correction measurement would actually degrade the analytical result.

2.3 The plasma converts metals in the sample from complex molecules to their atomic form and provides energy to excite them. Each metal will absorb

energy at a specific wavelength. Eventually the metal will return to its ground state by emitting the energy it absorbed. The ICP uses this emission of energy to quantify each metal. Each metal emits light at a different wavelength. This is called polychromatic. These wavelengths are separated by a monochromater and detected by a photomultiplier tube.

3.0 INTERFERENCES

3.1 Spectral interferences are caused by background emission from continuous or recombination phenomena, stray light from the line emission of high concentration elements, overlap of a spectral line from another element, or unresolved overlap of molecular band spectra.

3.1.1 Background emission and stray light are usually compensated for by subtracting the background emission determined by measurements adjacent to the analyte wavelength peak. Spectral scans of samples or single element solutions in the analyte regions may indicate when alternate wavelengths are desirable because of severe spectral interference. These scans will also show whether the most appropriate estimate of the background emission is provided by an interpolation from measurements on both sides of the wavelength peak or by measured emission on only one side. The locations selected for the measurement of background intensity will be determined by the complexity of the spectrum adjacent to the wavelength peak. The locations used for routine measurement must be free of off-line spectral interference (interelement or molecular) or adequately corrected to reflect the same change in background intensity as occurs at the wavelength peak. Spectral scans are performed whenever a new wavelength is being developed or when questions regarding a specific matrix arise.

3.1.2 Spectral overlaps are to be avoided by using an alternate wavelength or are compensated by equations that correct for interelement contributions. The ICP is capable of using equations for interelement correction. When utilized, it requires the interfering elements to be analyzed at the same time as the element of interest. When operative and uncorrected, interferences will produce false positive determinations and be reported as analyte concentrations. More extensive information on interferant effects at various wavelengths and resolutions is available in reference wavelength tables and books. The application of interelement correction factors is determined by testing the instrument with high concentration ranges of interfering analytes to compensate (off line or on line) for the effects of the interfering elements. Table 3 lists

potential spectral interferences determined on the ICP in our laboratory.

3.1.3 When using interelement correction factors, the interference is expressed as analyte concentration equivalents (i.e. false analyte concentrations) arising from a high concentration of the interference element in the LDR. Other concentrations of the interfering analyte cause relative readings (ie 10 mg/L of interference element would cause an interference about 1/10 as much as at 100 mg/L).

3.1.4 Interelement corrections are instrument specific. Any time a new instrument is placed into service, the interelement corrections must be established for that instrument. Selecting a background correction point where an interfering emission line may appear is avoided when practical. Interelement corrections that constitute a major portion of an emission signal may not yield accurate data. Analysts should not forget that some samples may contain uncommon elements that could contribute spectral interferences.

3.1.5 For each instrument, intensities will vary not only with optical resolution but also with operating conditions (such as power, viewing height and argon flow rate). The analyst is required to determine and document for each wavelength the effect from referenced interferences (Table 3) as well as any other suspected interferences that may be specific to the instrument or matrix. Whenever trace elements are determined, the analyst utilizes a computer routine for interelement corrections.

3.1.6 To determine the appropriate location for off-line background correction, the analyst scans the area on either side adjacent to the wavelength and records the apparent emission intensity from all other method analytes. This spectral information is documented and kept on file. A computer routine is used for automatic correction on all determinations. Tests to determine spectral interference are done using analyte concentrations that adequately describe the interference. The concentrations of interfering analytes used are found at the end of Table 3.

3.1.7 Absence of spectral interferences can be verified on samples of concern by scanning over a range of 0.174 nm (or width sufficient to view peak properly) centered on the wavelength of interest. This procedure is repeated when a new (unknown) matrix is analyzed. Background shift is corrected by the background correction points. An alternate wavelength that does not exhibit a background shift or spectral overlap may also be used.

3.1.8 Correction factors were tested on five consecutive days and found to be within the 20% criteria. Therefore on a weekly basis, whenever analysis is performed on metals that are affected by spectral interferences, the accuracy of interelement corrections is verified by analyzing the spectral interference check solution and

SIC blank. All interelement spectral correction factors are updated if the SIC solution and SIC blank do not consistently meet acceptance criteria. The standard solution should be inspected to ensure that there is no contamination that may be perceived as a spectral interference.

3.2 Physical interferences are effects associated with the sample nebulization and transport processes. Changes in viscosity and surface tension can cause significant inaccuracies, especially in samples containing high dissolved solids or high acid concentrations. Physical interferences are reduced by using a peristaltic pump and, if needed, by diluting the sample. An internal standard is added to all samples to further correct for physical interferences. Another problem that can occur with high dissolved solids is salt buildup at the tip of the nebulizer, affecting aerosol flow rate and causing instrumental drift. Samples with known high salts (ie. brines) are diluted during preparation to avoid salt buildup in the nebulizer.

3.3 Chemical interferences are minimized by careful selection of operating conditions, by matrix matching, and by standard addition procedures. Chemical interferences are highly dependent on matrix type and the specific analyte element.

3.4 Memory interferences are minimized by an appropriate rinse time with dilute acid between samples and standards. The instrument automatically increases the rinse time following a high sample.

1.1.1 Necessary rinse times for a particular element are estimated upon initial instrument set up. This is achieved by aspirating a standard containing elements at a concentration near the top of the linear dynamic range. The aspiration time for this sample should be the same as a normal sample analysis period, followed by analysis of the rinse blank at designated intervals. The length of time required to reduce analyte signals to *less than one half the reporting limit* is applied. If one half the reporting limit is less than the MDL, signals must be less than the MDL.

3.4.1 When there is suspected additional memory interference between samples, the rinse cycle is repeated and the sample is reanalyzed.

3.5 Analysts are aware that high salt concentrations can cause analyte signal suppressions and confuse interference tests. Our instrument does display negative values that would indicate the signal suppression.

4.0 APPARATUS AND MATERIALS

4.1 Inductively coupled argon plasma emission spectrometer: Perkin Elmer Optima 5300DV

- 4.1.1 Computer-controlled emission spectrometer with background correction.
- 4.1.2 Radio-frequency generator compliant with FCC regulations per PE manual.
- 4.1.3 Peristaltic pump.
  - 4.1.3.1 If an Elemental Science FAST system is used to deliver the standards and samples to the nebulizer then the variable speed peristaltic pump is required to deliver both carrier (i.e. rinse water) through the valve and internal standard directly to the nebulizer.
- 4.1.4 Autosampler.
- 4.1.5 Argon gas supply - high purity.

4.2 Labware - A clean laboratory work area designated for trace element sample handling must be used. Sample containers can introduce positive and negative errors in the determination of trace elements by contributing contaminants through surface desorption or leaching, depleting element concentrations through adsorption processes. All reusable labware (glass, quartz, polyethylene, PTFE, FEP, etc.) is sufficiently cleaned for the task objectives. Plastic volumetric flasks are rinsed with 1:1 nitirc and Nanopure water. See preparation SOP for washing instructions of digestion glassware.

- 4.2.1 Autopipets of suitable precision and accuracy

4.3 Plastic sample bottles: New bottles are used to store samples. Each new lot of bottles is tested for trace contaminants by analysis of an acid blank placed in one sample bottle from that lot. Results must be below the MDL.

5.0 REAGENTS AND STANDARDS

5.1 Reagent or trace metals grade chemicals shall be used in all tests. Unless otherwise indicated, it is intended that all reagents shall conform to the specifications of the Committee on Analytical Reagents of the American Chemical Society, where such specifications are available. Other grades may be used, provided it is first ascertained that the reagent is of sufficiently high purity to permit its use without lessening the accuracy of the determination. If the purity of a reagent is in question analyze for contamination. If the concentration of the contamination is less than the MDL then the reagent is acceptable.

- 5.1.1 Hydrochloric acid (conc), HCl.

5.1.2 Hydrochloric acid (1:1), HCl. Add 500 mL concentrated HCl to 400 mL water and dilute to 1 liter in an appropriately sized beaker.
5.1.3 Nitric acid (conc), HNO3.
5.1.4 Nitric acid (1:1), HNO3. Add 500 mL concentrated HNO3 to 400 mL water and dilute to 1 liter in an appropriately sized beaker.

5.2 Reagent Water. All references to water in the method refer to reagent water (Nanopure) unless otherwise specified. Reagent water will be interference free.

5.3 Standard Stock Solutions - Stock standards are purchased from a reputable supplier either separately or as mixed solutions. Two sources are acquired to verify instrument performance. Solutions are stored at room temperature. Manufacturer specified holding times are observed.

5.4 Working Standard Solutions – Solutions are made from above stock solutions in 1% HCl and 2% HNO_3. Solutions are stored at room temperature for up to six months. Solutions will be replaced sooner if comparison with QC samples indicate a problem. See standard log for recipes.

Note: *If the other QC standards indicate a problem with a mixed standard, each stock solution should be analyzed separately to determine the problem.* There can be slight high bias of Fe, Al, or other metals in mixed standards since stock sources of some metals contain trace amounts of other metals. If this bias causes QC samples to be out of range, each stock standard will be tested for the bias and this bias will be added to the true value of the metal in the standards.

5.5 Blanks - Four types of blanks are required for the analysis. The calibration blank is used in establishing the analytical curve, the laboratory reagent blank is used to assess possible contamination from the sample preparation procedure, the laboratory fortified blank is used to assess routine 1aboratory performance and a rinse blank is used to flush the instrument uptake system and nebulizer between standards, check solutions, and samples to reduce memory interferences.
5.5.1 The calibration blank for aqueous samples and extracts is prepared by acidifying reagent water to the same concentrations of the acids as used for the standards. The calibration blank is stored in a contaminant free plastic bottle.
5.5.2 The laboratory reagent blank (LRB) (method blank (MB)) must contain all the reagents in the same volumes as used in the processing of the samples. The LRB is carried through the same entire preparation scheme as the samples including sample digestion, when applicable.
5.5.3 The laboratory fortified blank (LFB) (Laboratory Control Sample (LCS)) is prepared by fortifying an aliquot of the laboratory reagent blank with all analytes to a suitable concentration

(approximately 100 times their respective MDL). 200 uL of an Intermediate standard is spiked into 5 mL of Nanopure water. See batch standard log for individual metal concentrations. The LFB must be carried through the same entire preparation scheme as the samples including sample digestion, when applicable.

5.5.4 The rinse blank is prepared by acidifying reagent water to 2% nitric acid/ 1% hydrochloric acid. (40mL nitric and 20mL hydrochloric into 2L of Nanopure)

5.6 Instrument Performance Check (IPC/CCV) Solution - The IPC is a mixed standard obtained from a reputable vendor. This standard is the same source as the calibration stock standards. The IPC solution is stored in a plastic bottle and analyzed as needed to meet data quality needs (Section 8.7.3). See standard log for recipe.

5.7 Initial Calibration Verification (ICV) – The ICV is a mixed standard obtained from a reputable vendor. This is a noncalibration source standard. The ICV solution is stored in a plastic bottle and analyzed as needed to meet data quality needs. See standard log for recipe.

5.8 Spectral Interference Check (SIC) Solutions - When interelement corrections are applied, SIC solutions are needed containing concentrations of the interfering elements at levels that will provide an adequate test of the correction factors. See standard log for preparation information.

5.9 Intensity Check (Plasma Solution) - The plasma solution is used for determining the optimum viewing position of the plasma above the work coil prior to using the method. The plasma solution (tuning solution) utilized on the Optima is 10 mg/L Manganese per manufacturers instruction PE CH8. The instrument is subjected prior to analysis to the autotune procedure provided with the software.

5.10 Internal Standard: A Lanthanum solution is added to all standards and samples. It is added either manually or through on line addition. See standard log for recipes.

6.0 SAMPLE COLLECTION, PRESERVATION, AND HANDLING

6.1 Prior to the collection of an aqueous sample, consideration should be given to the type of data required, (i.e., dissolved or total recoverable), so that appropriate preservation and pretreatment steps can be taken. If properly acid preserved, the sample can be held up to 6 months before analysis.

6.2 For the determination of the dissolved elements, the sample must be filtered through a 0.45-um pore diameter membrane filter at the time of collection or as soon thereafter as practically possible. (Glass or plastic filtering apparatus

are recommended to avoid possible contamination. Only plastic apparatus should be used when the determinations of boron and silica are critical.) Use a portion of the filtered sample to rinse the filter flask, discard this portion and collect the required volume of filtrate. Acidify the filtrate with (1+1) nitric acid immediately following filtration to pH < 2.

6.3 For the determination of total recoverable elements in aqueous samples, samples are not filtered, but acidified with (1+1) nitric acid to pH < 2 (normally, 3 mL of (1+1) acid per liter of sample is sufficient for most ambient and drinking water samples). Preservation may be done at the time of collection, however, to avoid the hazards of strong acids in the field, transport restrictions, and possible contamination it is recommended that the samples be returned to the laboratory within two weeks of collection and acid preserved upon receipt in the laboratory. Acid must be added to the original sampling container. Sample must be in contact with acid for at least 24 hours prior to removing an aliquot from the original container. If analysis begins <24 hours after sample preservation and pH verification, a qualifier will accompany the data.

6.4 The pH of all preserved samples is verified upon receipt by the laboratory, and if necessary, additional acid is added to bring the pH to <2. If the sample is reactive upon addition of acid, the splitter lets the reaction finish and then rechecks the pH.

6.5 For aqueous samples, a field blank is prepared and analyzed if required by the data user. Use the same container and acid as used in sample collection.

6.6 Samples must be digested prior to analysis.

6.7 Solid samples are collected unpreserved in glass jars or plastic bottles.

7.0 PROCEDURE

7.1 Preliminary treatment of most matrices is necessary because of the complexity and variability of sample matrices.

7.1.1 Samples must be digested prior to analysis. See SOP M02C for preparation of aqueous, solid, and sludge samples..

7.2 Calibration: Set up the instrument with proper operating parameters. The instrument must be allowed to become thermally stable before beginning (usually requiring at least 30 minutes of operation prior to calibration).

7.3 Profile and calibrate the instrument according to the instrument manufacturer's recommended procedures, using the typical mixed calibration standard solutions described in Step 5.4. Flush the system with the rinse blank (Step 5.5.3) between each standard or as the manufacturer recommends. (The average intensity of three readings for both standardization and sample analysis are used to reduce random error.) The calibration curve consists of a blank and a standard.

7.4 For all analytes and determinations, a ICV, CCV, and calibration blank is analyzed immediately following daily calibration. A CCV and calibration blank is analyzed after every tenth sample and at the end of the sample run. Analysis of the ICV, and CCV must verify that the instrument is within +/- 10% of calibration with relative standard deviation < 5% from replicate (minimum of two) integrations. If the calibration cannot be verified within the specified limits, reanalyze either the ICV or CCV (or both). If the second analysis confirms calibration to be outside the limits, the sample analysis is discontinued, the cause determined and the instrument recalibrated. All samples following the last acceptable ICV or CCV are reanalyzed. Data may be reprocessed under a new calibration using a later CCV and CCB as the calibrators. Reprocess all samples back thru the last working CCB and CCV. An earlier ICV and CCB reprocessed under the new calibration must work to accept the calibration. All data must be bracketed by working QC. The decision to accept any data when a CCV is out must be clearly documented and made by the QA Officer. The analysis data of the calibration blank, ICV, and CCV are kept on file with the sample analysis data.

7.5 The system is flushed with the rinse blank solution and then the next sample for at total of 1 minute before the analysis of each sample.

7.6 All data is sent to the LIMS.

7.6.1 Aqueous preparation initial and final volumes are 30/15mLs however since the La adjusts for this difference the default initial and final volumes in LIMS are set to 5/5mLs. If an aliquot other than the default volume is used during preparation, this variation is documented in the LIMS initial volume.

7.6.2 Nonaqueous samples initial and final volumes are defaulted into LIMS. Then a x2 dilution is made at the instrument which is entered into the DIL column (see section 7.6.3).

7.6.3 If a dilution is made at the instrument:

7.6.3.1 When using the Fast system, the dilution is entered in LIMS which will also adjust the sample result, MDL, and RL.

7.6.3.2 When not using the Fast system, the change in internal standard response will automatically adjust the result prior to data transfer. To correctly report the MDL and RL to reflect this dilution, the transferred result is divided by the dilution factor and entered as raw and the dilution factor is placed in the LIMS DIL column.

7.7 The result labeled in the run as “Si” is actually a silica (SiO_2) result and will be entered as such. Silicon standards used to calibrate for silica are entered as silica to account for this conversion.

7.8 Total Hardness may be calculated from the calcium and magnesium results as follows (SM 2340 B):

$$\frac{\text{Ca mg/L}}{20} + \frac{\text{Mg mg/L}}{12} \times 50 = \text{Total Hardness in mg eqiv. } CaCO_3/L$$

7.9 The MSA may be used if an interference is suspected or a new matrix is encountered. When the method of standard additions is used, standards are added at one or more levels to portions of a prepared sample. This technique compensates for enhancement or depression of an analyte signal by a matrix. It will not correct for additive interferences, such as contamination, interelement interferences, or baseline shifts. This technique is valid in the linear range when the interference effect is constant over the range, the added analyte responds the same as the endogenous analyte, and the signal is corrected for additive interferences. The simplest version of this technique is the single addition method. This procedure calls for two identical aliquots of the sample solution to be taken. To the first aliquot, a small volume of standard is added; while to the second aliquot, a volume of acid blank is added equal to the standard addition. The sample concentration is calculated by: multiplying the intensity value for the unfortified aliquot by the volume (Liters) and concentration (mg/L or mg/Kg) of the standard addition to make the numerator; the difference in intensities for the fortified sample and unfortified sample is multiplied by the volume (Liters) of the sample aliquot for the denominator. The quotient is the sample concentration. For more than one fortified portion of the prepared sample, linear regression analysis can be applied using a computer or calculator program to obtain the concentration of the sample solution.

7.10 The internal standard technique is utilized by our laboratory to check for interferences. Lanthanum is added to the samples, standards and blanks. The concentration of the internal standard must be equal in the standards, blank and samples. (See Standard Log for additional information on the standard preparation.) The element intensity is used by the instrument as an internal standard to ratio the analyte intensity signals for both calibration and quantitation. This technique is very useful in overcoming matrix interferences especially in high solids matrices. The analyst monitors internal standard response for large drops that might indicate improper spiking of La or sample matrix interference.

7.11 The upper limit of the range must be established by determining the signal responses from a minimum three, preferably five, different concentration standards across the range. One of these should be near the upper limit of the range. The upper range limit should be an observed signal no more than 10% below the level extrapolated from lower standards. Analyte concentrations above this upper range are diluted. For those analyses that are known interferences, and are present at above the linear range, the

analyst should ensure that the interelement correction has been applied correctly. New dynamic ranges should be determined whenever there is a significant change in instrument response.

7.12 A high check is performed with each run to prove linearity. Results must be within 90-110% recovery to accept sample with elevated results.

8.0 QUALITY CONIROL

Note: See also ESB SOP Q01 for general QC requirements

8.1 All quality control data are maintained and available for easy reference or inspection. See also ESB SOP Q01 for general QC requirements.

8.2 Demonstration of Method Capability

8.2.1 Before using this procedure to analyze samples, data documenting initial demonstration of performance is placed on file. The required data document the selection criteria of background correction points; analytical dynamic ranges, the applicable equations, and the upper limits of those ranges; the method and instrument detection limits; and the determination and verification of interelement correction factors or other routines for correcting spectral interferences. This data must be generated using the same instrument, operating conditions and calibration routine to be used for sample analysis. These documented data must be kept on file and be available for review by the data user or auditor.

8.2.2 Specific wavelengths are listed in Table 1. Other wavelengths may be substituted if they can provide the needed sensitivity and are corrected for spectral interference. Because of differences among various makes and models of spectrometers, specific instrument operating conditions cannot be provided. The instrument and operating conditions utilized for determination must be capable of providing data of acceptable quality to the program and data user. The analyst follows the instructions provided by the instrument manufacturer.

8.2.3 The plasma operating conditions need to be optimized prior to use of the instrument. The following procedure is recommended. The purpose of plasma optimization is to provide a maximum signal to background ratio for some of the least sensitive elements in the analytical array. The use of a mass flow controller to regulate the nebulizer gas flow rate greatly facilitates the procedure.

8.2.3.1 Profile the instrument to align it optically as it will be used during analysis. The following procedure can be

used for horizontal optimization. Aspirate a solution containing 1 mg/L of Manganese. Collect intensity data at the wavelength peak for the analyte at 1 mm intervals 15 mm above the load coil. (This region of the plasma is referred to as the analytical zone.)

8.2.3.2 The instrument operating condition finally selected as being optimum should provide the lowest reliable instrument detection limits and method detection limits.

8.2.3.3 If either the instrument operating conditions, such as incident power or nebulizer gas flow rate are changed, or a new torch injector tube with a different orifice internal diameter is installed, the plasma and viewing height should be re-optimized.

8.2.3.4 After completing the initial optimization of operating conditions, but before analyzing samples, an interelement spectral interference correction routine is established and initially verified to be used during sample analysis. A general description concerning spectral interference and the analytical requirements for background correction in particular are discussed in the section on interferences. Criteria for determining an interelement spectral interference is $\pm$ the method detection limit. Once established the entire routine must be periodically verified every six months.

8.2.4 Sensitivity, instrumental detection limit, precision, linear dynamic range, and interference effects are established for each individual analyte line utilized on each particular instrument. All measurements are required to be within the instrument linear range where the correction factors are valid.

8.2.4.1 Method detection limits are established for all matrices. An MDL study is completed whenever major equipment or procedural changes are made. Standards are spiked at a concentration of approximately *2*-5 times the estimated detection limit onto boiling stones for solids and into Nanopure water for ground waters. A minimum of seven replicates is analyzed. See QA Manual for calculation. MDL results must be below the reporting limit.

8.2.4.2 Determination of limits using reagent water or boiling stones blank represent a best case situation and do not represent possible matrix effects of real world samples.

8.2.4.3 If additional confirmation is desired, reanalyze the seven replicate aliquots on two or more nonconsecutive days and again calculate the method detection limit values for each day. An average of the three values for each analyte may provide for a more appropriate estimate. Successful analysis of samples with added

analytes or using method of standard additions can give confidence in the method detection limit values determined in reagent water.

8.2.4.4 All analytes concentrations determined must be within the linear calibration range of the analyte. Samples are diluted if readings exceed this established range.

Note: The analyst verifies that the instrument configuration and operating conditions satisfy the analytical requirements and maintains quality control data confirming instrument performance and analytical results.

8.3 Samples that exceed the linear calibration limit are diluted and reanalyzed.

8.4 A minimum of one method blank per sample batch is employed to determine if contamination or any memory effects are occurring. Results are reported per matrix type. For batch acceptance, MB values must be < ½ the reporting limit. If one half the reporting limit is less than the MDL, method blank values must be < the MDL for those analytes. Sample results at or above the reporting limit must be accompanied with a qualifier if the method blank exceeds a concentration greater than 1/10 the measured raw concentration of the sample. Blank results below the ½ the reporting limit (or below the MDL) are considered to be ND and will not require a note.

8.5 One Laboratory fortified blank (Blank Spike) is analyzed per batch per matrix type. Results must be within 85-115% recovery for the batch to be accepted.

8.6 At least one predigestion matrix spike/matrix spike duplicate are analyzed per matrix type per batch. Samples are chosen at random. See section 5.5.3 for recipe.

8.6.1 The relative percent difference between matrix duplicate determinations is to be calculated as follows:

$$RPD = \frac{|D(1) - D(2)|}{(D(1) + D(2))/2} \times 100$$

where:

RPD = relative percent difference.
D(1) = first sample value.
D(2) = second sample value (replicate).

8.6.2 (A control limit of +/- 20% RPD or within the documented historical acceptance limits for each matrix shall be used for sample values greater than ten times the instrument detection limit.)

8.6.3 The target range spiked sample or spiked duplicate sample recovery must be within 75-125% of the actual value.

8.7 Check the instrument standardization by analyzing appropriate QC samples as follows.

8.7.1 A calibration blank is analyzed immediately following daily calibration, after every 10 samples and at the end of the analytical run. The results of the calibration blank must be < *½ the reporting limit*. If one half the reporting limit is less than the MDL, calibration blank values must be < *the MDL* for those analytes. If this acceptance criteria is not met, terminate the analysis, correct the problem, recalibrate, and reanalyze the previous 10 samples. If the blank is less than 1/10 the concentration of the lowest sample, the analysis need not be terminated.

8.7.2 The ICV is a metals solution from a noncalibration source. Analyze the ICV immediately following initial calibration.

8.7.2.1 The ICV is used to verify initial calibration – acceptance criteria 10% recovery and the relative standard deviation of ICV replicate integrations must be <5%

8.7.2.2 If data must be reprocessed under a new calibration due to instrument drift later in the run, a later CCV may be used as the calibrator. The ICV must be processed under the new calibration to verify it - acceptance criteria: 10% recovery and the relative standard deviation of replicate integrations must be <5%.

8.7.3 The calibration is verified with the Calibration Verification Standard (CCV/IPC) immediately following daily calibration, after every 10 samples and at the end of the analytical run. It is made from the same source as the calibration standards.

8.7.3.1 The results of the calibration verification are to agree within 10% of the expected value and the relative standard deviation of CCV replicate integrations must be <5%; if not, terminate the analysis, correct the problem, and recalibrate the instrument.

8.8 Spectral interference check (SIC) solution - The laboratory verifies the interelement spectral interference correction routine by analyzing a SIC solution and blank weekly with a run that contains analytes of interest needing correction. The SIC solution and SIC blank are analyzed at the beginning of an analytical run (per 6010B 8.6.2). The interference check sample results must be within +/- 20% of the true value. The interference check blank must be less than $\pm$ the RL. All interelement spectral correction factors are updated if the SIC solution and SIC blank do not consistently meet acceptance criteria. To update correction factors separate standards of

each metal are analyzed at an elevated level within the LDR. Target metals are examined for any false response caused by the elevated metal.

8.9 A Detection Limit Check is analyzed with every calibration at the detection limit concentration. 50 – 150% of the expected value is considered acceptable.

8.10 Analyst Demonstration of Capability

8.10.1 Initial Demonstration: Prior to initial analysis of samples or when a significant change is made to the method, a Demonstration of Capability Study is performed by each analyst. This is accomplished by analysis of four replicates of a QC sample made at a concentration of 1-4 times the RL into Nanopure for aqueous studies* and onto boiling stones for nonaqueous studies. Concentrations of cation metals may be higher. Use sludge RL's for nonaqueous studies since they are lower than solid RL's. The average percent recovery of the QC samples must be 80-120% with a maximum %RSD of 20.

8.10.2 Demonstration of Continuing Proficiency: On an annual basis, each analyst must turn in valid LCS data from four consecutive batches or results from a successful Performance Evaluation Study. LCS percent recovery must meet laboratory prescribed acceptance criteria. Relative standard deviation between the four replicates must be less than or equal to 20%. Performance Evaluation Studies are performed twice a year.

*Former aqueous studies have been performed at 50-2000ppb.

8.11 LOD Verification: On a yearly basis, a QC sample is spiked at a level of not more than 1-4 times analyte MDL. The sample is analyzed. Analyte response must be greater than method blank response to verify the Level of Detection or MDL.

8.12 Corrective Action : For additional Information regarding corrective action, see SOP Q06 – Corrective Action

9.0 SAFETY: The toxicity or carcinogenicity of chemicals used in this method has not been precisely defined. Each chemical should be treated as a potential health hazard, and exposure to these chemicals should be minimized. A reference file of material data handling sheets is made available to all personnel involved in the chemical analysis.

9.1 See SOP S01 – Concentrated Acids and Bases
SOP S02 – Compressed Gas Cylinder Handling
SOP S03 – Spill Control Policy

10.0 Exporting and Entering Data: ICP-OPTIMA 5300DV

10.1 Delete calculations from QC and the source sample if not requested by the client i.e.: total hardness, total cations, sodium percentage,caco3, sar
10.2 Filtered blanks need to be qualified: Qbfilt
10.3 Add qualifier to source sample if the analyte is not requested: ?QC
10.4 Check for perfect zero - 0 could be saturated sample

10.5 Qualifiers:

- 10.5.1 Blank**:** A blank that does not meet the criteria must be qualified QBLK. A blank does not turn red if the result is too negative. The result may not be greater than +/- ½ the RL.
- 10.5.2 BS**:** A BS that is out of range may be qualified QLMS if the MS or MSD meet the BS criteria. If a BS is out of range and can not be qualified the analytes in question must be re-analyzed unless the results are ND and the BS is high.
- 10.5.3 MS, MSD:
  - 10.5.3.1 If both spikes are red check prep dilution and source sample prep dilution, correct on bench sheet if necessary.
  - 10.5.3.2 Check if source sample is greater than three times the BS value. If it is Qualify the MS, MSD: QM-3X
  - 10.5.3.3 If only one spike is out and the RPD is less than twenty and if there are ten or less analytes you can make the red spike the MSD and use the qualifier: QMSDr
  - 10.5.3.4 If all other analytes pass MS criteria and the LCS and CCVs pass for the out analyte, then sample matrix is suspected. Attach QMint to the QC and NMint to the sample and document fully.
  - 10.5.3.5 QFnt: If the source sample does not require the reporting of the analyte.
  - 10.5.3.6 QOcal if result is over LDR.
- 10.5.4 Verify dilutions on prep sheet and in LIMS
- 10.5.5 Samples, diluted, that result in an answer of ND or J flag**:** "N_RLm"
- 10.5.6 Right click Sort by analyte order
  - 10.5.6.1 Check ND results for any negatives greater than RL.
  - 10.5.6.2 If a blank has been qualified, check for any analytes less than 10 times the blank result. This result may be passable if it is ND, if it is not it must be RE'd and Qualified SUS; or qualified NBLK. The result may be reported if it is 10x greater than the blank result. The re's should be addressed now in Project Management, update status. Edit comment, create bench sheets if necessary.

10.5.6.3 Check for results over the LDR, these need to be RE'd , entered as 9999 and qualified ?SUS NOcal

10.5.7 Sort by Sample ID.

10.5.7.1 Calculate any sodium percentage or SAR's

10.5.8 Fill out peer review sheet

11.0 POLLUTION PREVENTION AND WASTE MANAGEMENT

11.1Waste Disposal

11.1.1 Instrument waste is neutralized and poured down the sink.

11.1.2 Sample waste after digestion is placed in the "High Acid Concentration Waste" barrel.

11.1.3 Samples that are hazardous (See Table 1 TTLC's and STLC's) are marked with a red dot on the original container. An entry is made in the log. Samples are held downstairs for pick up by an independent hazardous waste hauler

11.2 See SOP S05 – Neutralization Procedure for Acid and Alkaline Wastes
SOP S06 – Disposal of Chlorinated Solvents
SOP S07 – Pollution Prevention

12.0 METHOD PERFORMANCE

12.1 In an EPA round-robin Phase 1 study, seven laboratories applied the ICP technique to acid-distilled water matrices that had been spiked with various metal concentrates. Table 4 in EPA Method 6010B lists the true values, the mean reported values, and the mean percent relative standard deviations.

12.2 Performance data for aqueous solutions and solid samples from a multilaboratory study are provided in EPA Method 6010B, Tables 5 and 6.

13.0 DEFINITIONS

13.1See SOP Q15 – SOP Definitions

REFERENCES

EPA SW-846 Method 6010B revision 2, December 1996

Standard Methods For the Examination of Water and Wastewater APHA, AWWA, WEF 22nd Edition 2340 B.

Inductively coupled argon plasma emission spectrometer: Perkin Elmer Optima 5300DV Instrument Manual

Note: All *italicized items* are an indication of a variation from the method.

Approved by Julia Sudds Date 3/26/14

Table 3

		Interfering Metals																
Analyte	Wave-length	Al	Ba	Be	Ca	Cd	Co	Cr	Cu	Fe	Mg	Mn	Mo	Ni	Ti	Tl	V	Zn
Ag	328.07									x								
Al	396.15				x		x			x			x	x				x
As	188.98	x	x							x	x							
B	249.72						x			x			x					
Ba	233.53																x	
Be	313.11																x	
Cd	228.8						x											
Ca	317.93																	
Cd	228.80																	
Co	228.62		x												x			
Cr	267.72																	
Cu	327.39				x	x	x			x			x		x		x	
Fe	238.2						x				x							
K	766.49																	
Mg	285.21																	
Mn	257.61												x					
Mo	202.03																	
Na	589.59																	
Ni	231.6		x				x											
Pb	220.35	x			x				x									
Sb	206.84				x			x		x			x					
Se	196.03	x			x		x	x		x			x	x	x	x	x	
Tot Si	251.61												x					
Sn	189.93									x					x			
Sr	407.77		x	x						x	x	x		x				
Ti	334.94																	
Tl	190.8						x				x	x	x		x		x	
V	310.23	x	x	x	x		x	x		x	x	x			x			x
Zn	206.2	x	x					x		x	x	x	x					

x - This metal interferes with this analyte. Spectral correction must be applied.