MARYLAND DEPARTMENT OF NATURAL RESOURCES

SECTION 106
AMBIENT WATER QUALITY MONITORING
(CORE/TREND MONITORING)

QUALITY ASSURANCE PROJECT PLAN

September 15, 2015

MARYLAND DEPARTMENT OF NATURAL RESOURCES

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The Maryland Department of Natural Resources (DNR) seeks to preserve, protect and enhance the living resources of the state. Working in partnership with the citizens of Maryland, this worthwhile goal will become a reality. This publication provides information that will increase your understanding of how DNR strives to reach that goal through its many diverse programs.

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Maryland Department of Natural Resources
Section 106 - Ambient Water Quality Monitoring
(Core/Trend Monitoring)
Quality Assurance Project Plan
July 1, 2015 – June 30, 2016

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ACRONYMS AND ABBREVIATIONS

C	Carbon
CBP	EPA's Chesapeake Bay Program
CBPO	EPA's Chesapeake Bay Program Office
CBL	University of Maryland's Chesapeake Biological Laboratory
cm	Centimeter
CSSP	Coordinated Split Sample Program
DIWG	Data Integrity Workgroup (a workgroup of the Chesapeake Bay Program, formerly AMQAW)
DHMH	Maryland Department of Health and Mental Hygiene
MDNR	Maryland Department of Natural Resources
DO	Dissolved oxygen
DOC	Dissolved organic carbon
EPA	U.S. Environmental Protection Agency
g	Gram
H_2O	Dihydrogen oxide (water)
L	Liter
m	Meter
MDE	Maryland Department of the Environment
min.	Minute
mg	Milligram
ml	Milliliter
mm	Millimeter
N	Nitrogen
NIST	National Institute of Science and Technology
NO_2	Nitrite
$NO_{2,3}$	Nitrate + nitrite
NO_3	Nitrate
P	Phosphorus
PC	Particulate carbon
PN	Particulate nitrogen
PO_4	Phosphate
PP	Particulate phosphorus
QAO	Quality Assurance Officer (unless otherwise noted, this refers to the DNR QAO)
QAPP	Quality Assurance Project Plan
RP	Replicate
TDN	Total dissolved nitrogen
TDP	Total dissolved phosphorus
TSS	Total suspended solids
USGS	U.S. Geological Survey
^oC	Degrees Celsius

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PROJECT MANAGEMENT

A1 Project/Task Organization

This section lists the individuals responsible for the major aspects of Maryland's Ambient Water Quality Monitoring Program. The flow of project tasks is indicated in Figure 1.

Director and Principal Investigator: Bruce Michael, Resource Assessment Service, MDNR. 410-260-8627, bruce.michael@maryland.gov

Responsibilities: The director and principal investigator is responsible for overseeing the administrative aspects of the program including fiscal management, coordination among other DNR managers and coordination with cooperating agencies and institutions. This individual is also responsible for the technical design, management and data analysis of the program.

Quality Assurance Officer: Bruce Michael, Resource Assessment Service, MDNR. 410-260-8627, bruce.michael@maryland.gov

Responsibilities: The quality assurance officer is responsible for documenting and assuring the completion of field, laboratory and data management procedures that comprise this study.

Field Sampling Operations: Kristen Heyer, Monitoring and Non-tidal Assessment Division, MDNR. 410-990-4600, kristen.heyer@maryland.gov and Laura Fabian, Monitoring and Non-tidal Assessment Division, MDNR. 410-990-4524, laura.fabian@maryland.gov

Responsibilities: These individuals are responsible for administration of the field sampling activities including sample collection, sample storage and sample delivery to laboratories.

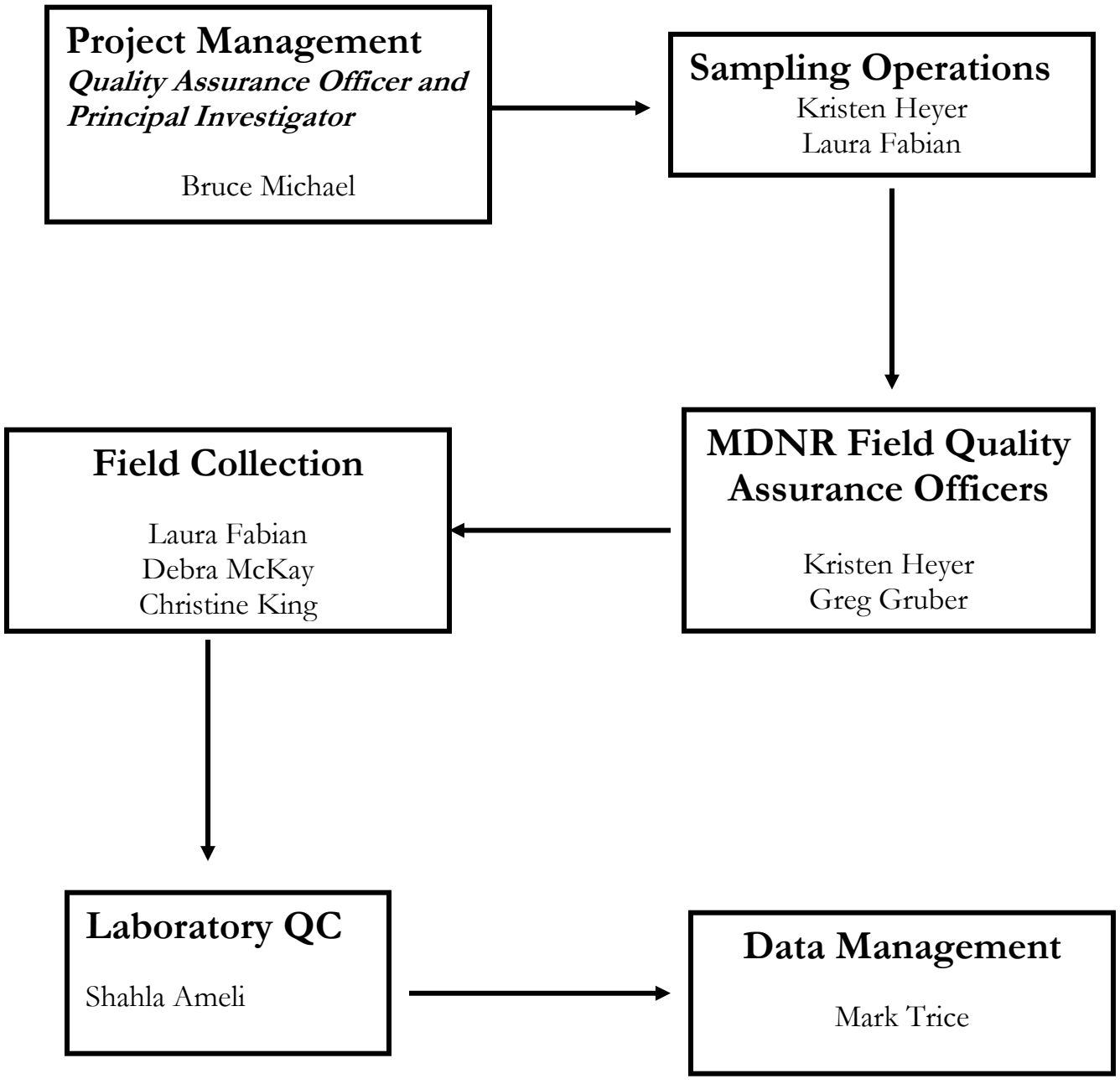
Laboratory Analyses/Water Column Chemistry: Shahla Ameli, Supervisor, Division of Environmental Chemistry, DHMH. 443-681-3855, shahla.ameli@maryland.gov

Responsibilities: This person oversees the laboratory that does all of the nutrient analysis and water chemistry for the Ambient Water Quality Monitoring Program.

Data Management; Mark Trice, Program Chief of Water Quality Informatics, Tidal Ecosystem Assessment, MDNR. 410-260-8649, mark.trice@maryland.gov

Responsibilities: This person is responsible for overseeing the management of field and laboratory data collected under this program; managing historical field and laboratory data collected under this program; and maintaining existing data management software.

Figure 1. Project Organization for Maryland's 106 Ambient Water Quality Monitoring Program



A2 Problem Definition/Background

Maryland DNR's Ambient Water Quality Monitoring Program (Core/Trend Monitoring) is part of a nationwide ambient monitoring effort designed to measure progress towards achieving EPA's national water quality goals. This program was initiated in 1974 to meet an EPA-mandated monitoring requirement for the State of Maryland to collect data that can be used to detect status and trends in the quality of the State's waters. To detect status and trends in Maryland's waters, ambient fixed stations were located in water use areas, problem areas, land use areas and in areas where future development may impact water quality. As of 1 July 1995, responsibility for Maryland's Ambient Water Quality Monitoring Program was transferred from the Maryland Department of the Environment to the Maryland Department of Natural Resources.

A3 Project/Task Description

Maryland's portion of this national ambient monitoring effort includes 37 Core stations located in non-tidal and tidal freshwater and estuarine areas and 25 Trend stations located on larger, non-tidal streams and rivers (4^{th} order and larger). A map of station locations is presented in Figure 2 and a description of each station is presented in Table 1. The 62 stations that comprise this monitoring program are sampled monthly, year-round, for physical and chemical parameters.

A3.1 Core Stations

Core Station selection was based upon EPA's Basic Water Monitoring Program (BWMP) manual (EPA 1976). The selection of stations for the Core network was guided primarily by the need to assess conditions in water use areas. These included recreational areas, surface water supply areas, land use areas and potential areas of development. Since these data are used in the national assessment program, both impacted and non-impacted areas were included in the network to ensure that the evaluation would not be biased. Sampling stations were also selected to be representative of various regions in Maryland. Where consistent with the aforementioned criteria, stations were located to maintain continuity with existing sites. Overall, the locations of the 37 stations cover the major freshwater rivers of the State that flow into the Chesapeake Bay and also bracket major population centers.

Water quality data are collected monthly from each of the 37 Core stations located throughout the State. Surface samples are collected at 29 freshwater Core stations, but at various depths at the 8 estuarine stations. Nine sampling stations in the western part of the State (located in the Youghiogheny, North Branch Potomac, or Upper Potomac River) are sampled for additional water quality parameters (sulfate and chloride) to monitor for the impacts of acid mine-drainage. The sulfate and chloride sampling is described in the scope of work for the 106 supplemental memorandum of understanding between MDE and DNR. For logistical reasons, samples for the 8 estuarine Core stations are collected during sampling for other Maryland DNR monitoring programs. Samples for stations CB2.1, CB3.3C, and CB5.1 are collected during Bay mainstem cruises, while ET5.2, XGG 8251, WT5.1, RET2.4 and TF2.3 are collected during tributary sampling (see Figure 2, estuarine Core stations are presented in orange). Sampling protocols for these 8 stations are outlined in this Quality Assurance Project Plan (QAPP). Sample analysis is conducted by the University of Maryland, Chesapeake Biological Laboratory, Nutrient Analytical Services Laboratory

(NASL). The analytical methods utilized by NASL are detailed in Appendix VII of the Quality Assurance Project Plan for Maryland DNR's Chesapeake Bay Water Quality Monitoring Program – Chemical and Physical Properties Component. The latest version of this Plan is available at: http://mddnr.chesapeakebay.net/eyesonthebay/documents/MdDNR_MTQAPP2014.pdf.

Thirteen stations (identified with an asterisk in Table 1) are also sampled separately as part of the Chesapeake Bay Program's Non-tidal Network (117(d)). For additional information on this monitoring program, please see Maryland DNR's Non-tidal Network, Quality Assurance Project Plan. The latest version of this QAPP is available at: http://mddnr.chesapeakebay.net/eyesonthebay/documents/MD117dQAPP1415.pdf

A3.2 Trend Stations

In addition to the Core stations, water samples are also collected monthly, year-round, at an additional 25 freshwater Trend stations. These stations are sampled to enhance the amount of water quality information available for status and trends assessments in the freshwater portions of Maryland's larger streams and rivers.

Both Core and Trend water quality samples are collected and analyzed in the exact same manner and follow the same quality assurance and quality control protocols. Parameters and analytical methods are listed in Table 2.

A3.3 Schedule of Tasks and Projects

Maryland DNR's Ambient Water Quality Monitoring Program was initiated in its present form. Since this is an ongoing effort, the schedule and time required to complete each of the tasks (from sampling to data verification) associated with monthly collections are presented below.

Schedule for Monthly Sampling and Data Processing

Task	Time Required	Cumulative Time
Sample Collection	20 person days	3 weeks
Laboratory Analysis	1 week	4 weeks
Data Verification	1 week	5 weeks
Data Keypunching	1 week	6 weeks
Final Data Verification	2 weeks	8 weeks

Data analysis to determine status and trends in water quality are calculated after the data has undergone the quality assurance process. Status and trends are calculated annually. Appendix I provides a complete description on how status and trends are calculated. Results will continue to be provided on the Internet at (http://www.dnr.maryland.gov/streams/) as well as provided directly to Tributary Strategy workgroups. The primary reporting mechanism for this program is the State's integrated 305b/303d report. This report provides a comprehensive assessment of Maryland's waters incorporating MDNR's Ambient Water Quality Monitoring Program results with other intensive or routine water quality surveys within Maryland. The integrated report is submitted to EPA's regional office for review and approval. Draft and final versions are posted on Maryland Department of the

Environment's website (www.mde.state.md.us) for access by federal, state, and local agencies as well as other officials and the public.

A4 Quality Objectives and Criteria

Maryland DNR's Ambient Water Quality Monitoring Program is designed to provide laboratory and field data that will help the State of Maryland to detect status and trends in the quality of its waters. Assessments of the quality of the data collected through this program can be expressed in terms of representativeness, comparability, completeness, accuracy and precision.

A4.1 Representativeness

Representativeness is the degree to which the sample data represent the actual conditions or concentrations present in the sampled population or area. Representativeness can be affected by experimental design and sample collection and handling. The experimental design (described in section A6) for this monitoring program requires monthly sampling (12 collections/year) for physical and chemical parameters on a pre-determined date, which is adequate for capturing long-term annual trends in concentration (Alden et al. 1994).

A4.2 Comparability

Comparability refers to the confidence with which one data set can be compared with another. Comparability must be ensured so that the results for one station are of comparable quality to other stations. In addition, the data generated by Maryland's Ambient Water Quality Monitoring Program must also be of comparable quality to the data generated by other states and laboratories. Comparability among data sets is assured through the use of consistent field methods and protocols, participation in the Data Integrity Workgroup and the use of field splits and blind audit samples. Comparability of monitoring data is achieved as a result of quality assurance procedures at each phase of data gathering and processing. It includes representative sampling and sample handling procedures, uniform laboratory methods and validation of laboratory data and procedures for reduction, validation and reporting of environmental data.

A4.3 Completeness

Completeness is a measure of the amount of valid data obtained compared to the amount that was expected under normal conditions. Completeness is a condition to be achieved in order to meet the data requirements of the program. Factors that can affect completeness include problems encountered by the field crews such as adverse weather conditions or equipment failures and laboratory-related issues such as sample preservation, exceeding holding times and accidents. To ensure that data are of the quality required to aid and support management decisions, Maryland's Ambient Water Quality Monitoring Program strives to provide monitoring data of known and consistent quality by generally following the guidelines outlined in Section E of the Recommended Guidelines for Sampling and Analysis in the Chesapeake Bay Monitoring Program, August 1996 (EPA 1996). These guidelines recommend precision goals of field and laboratory measurements of < 20 percent of the coefficient of variation; accuracy goals within 80 to 120 percent, and the completeness

goals of 90 percent.

A4.4 Accuracy

The accuracy (closeness to the true value) of the collected data is controlled and assured by the proper use, calibration, and maintenance of both field and laboratory equipment for the measurement of physical and chemical parameters. All instruments are identified by a unique number, used as an index for documentation of calibration, repair, and preventative maintenance. Multiparameter field instruments are calibrated prior to field sampling to ensure accuracy. Where possible, standards used for calibration purposes are validated against a primary standard such as those available from the National Institute of Science and Technology (NIST). Daily quality control checks (including the running of blanks and standards) are used to control and assure laboratory accuracy.

Accuracy of laboratory results is also assessed through MDNR's participation in the Chesapeake Bay Coordinated Split Sample Program (CSSP), a split sampling program in which nine laboratories involved in Chesapeake Bay monitoring analyze quarterly, coordinated split samples. CSSP was established in June 1989 to establish a measure of comparability between sampling and analytical operations for water quality monitoring throughout the Chesapeake Bay and its tributaries. MDNR follows the protocols in the Chesapeake Bay Coordinated Split Sample Program Implementation Guidelines (EPA 1991) and its revisions. Split samples are collected quarterly. Results are analyzed by appropriate statistical methods to determine if results differ significantly among laboratories. When a difference occurs, discussion begins regarding techniques and potential methods changes to resolve discrepancies and identify potential problems. Additionally, DHMH participates biannually in the USGS reference sample program and permits USGS to release the results to the Chesapeake Bay Program Quality Assurance Officer. Laboratory accuracy is 90-110% recovery.

A4.5 Precision

Precision (repeatability) measures the closeness of values for a parameter within a data set. Quality control samples along with appropriate statistical techniques are used to ensure precision in the production of laboratory data. Precision of the chemical analytical methods is determined and documented from duplicate analyses. DHMH performs precision calculations for laboratory duplicates. Every tenth sample is analyzed in duplicate. The acceptable value for the relative percent difference (RPD) is +/- 10%. If the calculated RPD does not fall within the acceptable range, the corresponding analysis is repeated. Precision of the entire measurement system for laboratory-analyzed parameters, including field processing, storage, and chemical analysis, can be assessed from duplicate field samples. Maryland DNR data analysts responsible for quality assurance checks examine field duplicate data. Based on preliminary analysis, there can be relatively large differences in measured values for certain water quality parameters. Maryland DNR's data management section is currently devising more robust procedures to address this issue in a statistically quantifiable manner.

A5 Special Training/Certification

Maryland DNR field personnel are required to demonstrate proficiency in all aspects of sample collection to the Chesapeake Bay Program Quality Assurance Coordinator, who conducts annual site visits to assure the continued proficiency of field personnel and adherence to the procedures specified in the Department's SOP. Training for field personnel who collect samples for the Ambient Water Quality Monitoring Program is provided by experienced field staff and the field Quality Assurance Officers (Greg Gruber and Kristen Heyer). All training and procedures adhere to the Standard Operating Procedures developed for the Ambient Water Quality Monitoring Program.

Like DNR field personnel, analysts working for DHMH are required to demonstrate proficiency in laboratory procedures. New analysts are trained by an experienced analyst on the laboratory procedures he/she will be assigned to perform. Training is documented using the Division's training forms and signed by the Supervisor, Quality Assurance Officer and the Division Chief.

A6 Documentation and Records

Documenting sampling events is an important component of the Ambient Water Quality Monitoring Program. Field crews document all data obtained in the field on field sheets. Examples of the field sheets are provided in the Standard Operating Procedures #PR-03: Maryland Core/Trend Monitoring Program (Appendix II). Since the data generated by this program are not used for legal purposes, a formal chain-of-custody sheet is not required. Field sheets and any information regarding a specific problem and/or event during a sampling run, as well as modifications to the sampling program are maintained by MDNR field office staff.

A water quality monitoring field sheet is completed on arrival at each station. This form is used to record field measurements and other data pertinent to the collection of samples, including sample type and date and time of collection. An instrument specific calibration logbook is also used to record instrument calibration data, notes on instrument testing, and notes on instrument performance, problems and repairs.

Critical project personnel receive copies of the QAPP (please see distribution list on page vii). The QAPP and all associated SOPs referenced herein will be updated annually by June 30th. Any modifications to the QAPP will be reviewed and approved by personnel conducting the sampling, the principal investigators and MDNR's Quality Assurance Officer. Once approved, the final version will be made available to all interested parties by placing downloadable copies on the CBP's and MDNR's websites. Project reporting to management will be accomplished by quarterly progress reports of activities. Electronic summaries of provisional instantaneous water quality data will be provided on an annual basis. The final data set is stored in a local designated DNR database directory for data user access. Data requests should be directed to Mark Trice, Program Chief of Water Quality Informatics (410-260-8630). Data are also entered into the CIMS database and are available to the public via the Bay Program web site at http://www.chesapeakebay.net/data.

Figure 2. Maryland Department of Natural Resource's Ambient Water Quality Monitoring Stations

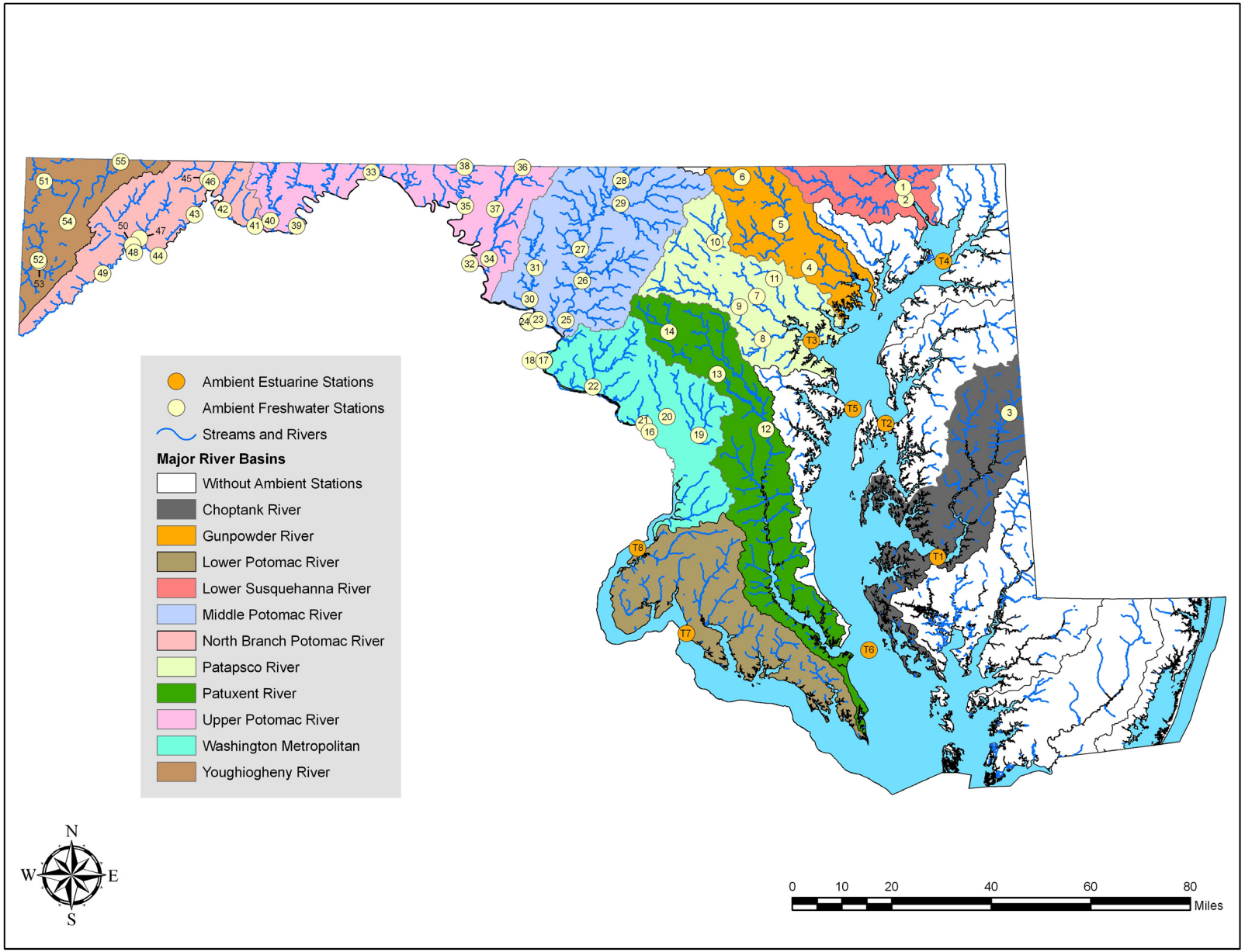


Table 1. Maryland Department of Natural Resource's Ambient Water Quality Monitoring Station Information.

Map #	Station I.D.	Stream Name	River Mile	Longitude (NAD 83)	Latitude (NAD 83)	Description and Site Type (Core or Trend)
02-12-02 SUSQUEHANNA RIVER BASIN						
1	CB1.0 (SUS 0109)	Susquehanna River	10.90	076 10.5023788	39 39.3729986	Below Conowingo Dam at boat ramp - gaging station USGS-01578310 - C
2	DER 0015*	Deer Creek	1.50	076 09.8863318	39 37.4085651	Bridge on Stafford Bridge Road – USGS-01580520- Tr
02-13-04 CHOPTANK RIVER BASIN						
T1	ET5.2 (XEH 4766)	Choptank River		076 03.5202530	38 34.8394323	At drawspan on U.S. Rt. 50 bridge - C
3	ET5.0 (CHO 0626)	Choptank River	62.60	075 47.1864631	38 59.8311087	At Red Bridges near Sewell Mills USGS-01491000 - C
02-13-05 CHESTER RIVER BASIN						
T2	XGG 8251	Kent Island Narrows		076 14.8401240	38 58.2675736	At drawspan on MD Route 18 bridge - C
02-13-08 GUNPOWDER RIVER BASIN						
4	GUN 0125	Gunpowder Falls	12.50	076 31.7336277	39 25.5375149	At bridge on Cromwell Bridge Road - C
5	GUN 0258*	Gunpowder Falls	25.80	076 38.1520258	39 33.0386351	End of Glencoe Road at old bridge crossing USGS – 01582500 - C
6	GUN 0476	Gunpowder Falls	47.60	076 46.8285205	39 41.3615564	Bridge at Gunpowder Road –USGS-01581810- C
02-13-09 PATAPSCO RIVER BASIN						
7	GWN 0115*	Gwynns Falls	11.50	076 43.5833003	39 20.5671785	At bridge on Essex Road in Villa Nova near gaging station USGS-01589300 - C
8	PAT 0176	Patapsco River	17.60	076 42.3202382	39 13.0687759	At bridge on Washington Boulevard (U.S. Rt. 1) - C

Table 1. Maryland DNR's Ambient Water Quality Monitoring Sampling Locations (cont.)

Map #	Station I.D.	Stream Name	River Mile	Longitude (NAD 83)	Latitude (NAD 83)	Description and Site Type (Core or Trend)
02-13-09 PATAPSCO RIVER BASIN (cont.)						
9	PAT 0285	Patapsco River	28.50	076 47.5345192	39 18.7467204	At bridge on Md. Rt. 99 near Hollofield gage USGS-01589000 - Tr
10	NPA 0165*	North Branch Patapsco River	16.50	076 52.9250807	39 28.9671330	Wire pedestrian bridge in industrial plant near gage USGS-01586000 - C
11	JON 0184	Jones Falls	10.8	076 39.68155	39 23.0730508	Bridge on Falls Road (Md. Rt. 25) near Sorrento gage– USGS-01589440 - C
T3	WT5.1 (XIE 2885)	Patapsco River	5.31	076 31.3521434	39 12.7856735	At buoy 5M, Hawkins Point - C
02-13-11 PATUXENT RIVER BASIN						
12	TF1.0 (PXT 0603)	Patuxent River	61.58	076 41.6465749	38 57.3343692	At bridge on U.S. Route 50 USGS-01594440 - C
13	PXT 0809	Patuxent River	81.91	076 52.4958913	39 07.0081428	At the gaging station just below Rocky Gorge Dam USGS-01592500 - C
14	PXT 0972*	Patuxent River	102.22	077 03.3713472	39 14.3584868	At bridge on Md. Route 97 near Unity gage USGS-01591000 - C
02-13-99 CHESAPEAKE BAY MAINSTEM						
T4	CB2.1 (XJH 6680)	Chesapeake Bay		076 01.5594740	39 26.4894865	200 yds. northeast of buoy RBA, mid-bay, south of Turkey Point, 15' depth - C
T5	CB3.3C (XHF 1373)	Chesapeake Bay		076 22.1808	39 00.84772	2100 yds., NE of Sandy Point, 55' depth - C
T6	CB5.1 (XCG 8613)	Chesapeake Bay	94	076 18.6833820	38 18.6510555	Off Patuxent River near mid-channel - depth 55 - 100' - C
02-14-01 LOWER POTOMAC RIVER BASIN						
T7	RET2.4 [a] (XDC 1706)	Potomac River		076 59.4376865	38 21.7559638	In mid-channel at Morgantown Bridge (U.S. Route 301), 58' depth - C
T8	TF2.3 [a] (XEA 6596)	Potomac River		077 10.4383095	38 31.8040859	Buoy N54 off Indian Head, 44' depth - C

Table 1. Maryland DNR's Ambient Water Quality Monitoring Sampling Locations (cont.)

Map #	Station I.D.	Stream Name	River Mile	Longitude (NAD 83)	Latitude (NAD 83)	Description and Site Type (Core or Trend)
02-14-02 WASHINGTON METROPOLITAN AREA						
16	POT 1184 [a]	Potomac River	118.40	077 07.6400929	38 56.8928182	At gaging station just above Little Falls Dam USGS-01646500 - C
17	POT 1471 [a]	Potomac River	147.10	077 31.2750641	39 09.2651668	At Eastern Terminus off Whites Ferry - C
18	POT 1472 [a]	Potomac River	147.0	077 31.3390209	39 09.3307768	At Western Terminus of Whites Ferry - Tr
19	ANA 0082 [a]	Anacostia River	8.20	076 56.6068030	38 56.3360716	At boat ramp in Bladensburg Waterfront Park - C
20	RCM 0111 [a]	Rock Creek	11.10	077 03.7817405	38 59.5812919	At bridge on Md. Route 410 - Tr
21	CJB 0005 [a]	Cabin John Branch	0.50	077 08.9301668	38 58.4069338	At bridge on MacArthur Boulevard - Tr
22	SEN 0008 [a]	Seneca Creek	0.80	077 20.3781583	39 04.7749739	At bridge on Md. Route 112 - Tr
02-14-05 UPPER POTOMAC BASIN						
23	POT 1595 [a]	Potomac River	159.50	077 32.6203211	39 16.4085768	At boat ramp just downstream of MD side of U.S. Rt. 15 near Pt. of Rocks USGS-01638500 – Tr
24	POT 1596 [a]	Potomac River	159.55	077 32.8740048	39 163250283	At boat ramp just upstream of VA side of U.S. Rt. 15 near Pt. of Rocks - Tr
25	MON 0020 [a]	Monocacy River	2.00	077 26.4946321	39 16.3025469	Bridge on MD 28 - C
26	MON 0155 [a]	Monocacy River	15.50	077 22.8656221	39 23.2669471	Pine Cliff Park ramp upstream of bridge - C
27	MON 0269 [a]	Monocacy River	26.90	077 23.3631412	39 28.8165566	Bridge on Biggs Ford Rd. - C
28	MON 0528* [a]	Monocacy River	52.80	077 14.0929806	39 40.7500155	At bridge on MD 140, near gage house in Bridgeport– USGS – 1639000 - C
29	BPC 0035 [a]	Big Pipe Creek	3.50	077 14.2924934	39 36.7306812	Bridge on Md. Rt. 194 USGS gaging station USGS – 1639500 - Tr

Table 1. Maryland DNR's Ambient Water Quality Monitoring Sampling Locations (cont.)

Map #	Station I.D.	Stream Name	River Mile	Longitude	Latitude	Description and Site Type (Core or Trend)
02-14-05 UPPER POTOMAC BASIN (cont.)						
30	CAC 0031 [a]	Catoctin Creek	3.10	077 34.8107379	39 19.9069327	Right bank, just upstream of bridge on Md. Route 464 - Tr
31	CAC 0148* [a]	Catoctin Creek	14.80	077 33.5401108	39 25.5468858	Right bank just downstream ofbridge on Md. Route 17, at gaging station USGS-01637500 - Tr
32	POT 1830 [a]	Potomac River	183.00	077 48.1594887	39 26.1046394	At boat ramp WV side below bridge on Md. Rt. 34, near discontinued gage USGS-01618000 (discontinued 2004) - C
33	POT 2386	Potomac River	238.60	078 10.5781510	39 41.4671425	At rocky outcropping downstream of boat ramp @ C&O park, near gaging station 0.5 mile below bridge on U.S. Rt. 522 USGS-0161300 - C
34	ANT 0044* [a]	Antietam Creek	4.40	077 43.8991688	39 27.0219634	Downstream of Sharpsburg gage house, streamside – USGS-01619500 - Tr
35	CON 0005	Conococheague Creek	0.50	077 49.2963323	39 36.1943845	Md. 68 bridge - C
36	ANT 0366*	Antietam Creek	20.30	077 36.4935486	39 42.9592863	At bridge on Millers Church Road adjacent to gaging station at Rocky Forge – USGS – 01619000 - Tr
37	ANT 0203	Antietam Creek	20.30	077 42.6475848	39 35.6775584	At bridge on Poffenberger Rd. near Funkstown - C
38	CON 0180*	Conococheague Creek	18.00	077 49.5032338	39 42.9627173	At gaging station on Wishard Rd. USGS-01614500 - Tr
39	POT 2766	Potomac River	276.60	078 27.2695565	39 32.3189316	At bridge on Md. Rt. 51 near Paw Paw, W. Va. USGS-01610000 - Tr
40	TOW 0030* [+]	Town Creek	3.00	078 33.2032866	39 33.1821660	At gage on Pack Horse Road USGS-01609000 - C
02-14-10 NORTH BRANCH POTOMAC RIVER BASIN						
41	NBP 0023 [+]	North Branch Potomac	2.30	079 39.3300605	37 58.4618290	Toll bridge at Oldtown - Tr
42	NBP 0103 [+]	North Branch Potomac	10.30	078 43.8873501	39 34.9607011	Boat ramp off Rt. 51 in Spring Gap - C

Table 1. Maryland DNR's Ambient Water Quality Monitoring Sampling Locations (cont.)

Map #	Station I.D.	Stream Name	River Mile	Longitude	Latitude	Description and Site Type (Core or Trend)
02-14-10 NORTH BRANCH POTOMAC RIVER BASIN (cont.)						
43	NBP 0326	North Branch Potomac	32.60	078 50.3348823	39 34.0064182	Bank sample upstream MD route 936 bridge at Pinto- USGS:01600000 (discontinued) - C
44	NBP 0461 +	North Branch Potomac	46.10	078 58.3048527	39 26.6943955	Bank sample under U.S. Route 220 bridge- Tr
45	BDK 0000	Braddock Run	0.01	078 47.4487205	39 40.2286587	Old Mt. Savage Road bridge - Tr
46	WIL 0013*	Wills Creek	1.38	078 46.8174564	39 39.7110428	Locust Grove Road bridge crossing near gaging station USGS-01601500 - Tr
47	GEO 0009* +	Georges Creek	0.90	079 02.6819423	39 29.6183080	Victory bridge in Westernport next to Town Hall - near USGS – 01599000 - C
48	NBP 0534 +	North Branch Potomac	53.48	079 04.0814362	39 28.7536221	North Branch at Bloomington just upstream of confluence with Savage River USGS-01596000 - C
49	NBP 0689	North Branch Potomac	68.90	079 10.7614696	39 23.3607386	Rt. 38 bridge over North Branch USGS – 01595500 - C
50	SAV 0000	Savage River	0.02	079 04.0838436	39 28.8359583	Savage River at Md. 135 - Tr
05-02-02 YOUGHIOGHENY RIVER BASIN						
51	YOU 0925 +	Youghiogheny River	94.00	079 24.5074447	39 39.1739972	Bridge Crossing in Friendsville on Main street near USGS – 03076500 - Tr
52	YOU 1139	Youghiogheny River	115.91	079 25.3143438	39 25.4158776	Liberty Street/Herrington Manor Bridge crossing near USGS – 03075500 - Tr
53	LYO 0004 +	Little Youghiogheny R.	0.38	079 25.1550365	39 25.1060846	Bridge Crossing at Oakland/Rosedale Rd. - Tr
54	CCR 0001 +	Cherry Creek	0.15	079 18.9509986	39 32.2335135	Bridge Crossing on State Park Road – USGS-03075905- Tr
55	CAS 0479	Casselman River	47.92	079 08.1846184	39 42.1242778	Casselman River where crossed by River Road at USGS – 03078000 - Tr

* Ambient Water Quality Monitoring Stations that are also sampled separately as part of the Chesapeake Bay Program's Non-tidal Network – 117(d). For additional information on this monitoring program, please see Maryland DNR's Non-tidal Network, Quality Assurance Project Plan, 2014.

+ Western Maryland stations sampled for additional water quality parameters - sulfate and chloride (106 supplemental)

[a] Potomac River Basin Ambient Water Quality Monitoring Stations sampled for an additional water quality parameter – 5-day biochemical oxygen demand

( ) = Original Station Name

Table 2. Water Quality Parameters, Methods, Preservation/Holding Times and Method Detection Limits for MDNR's Ambient Water Quality Monitoring Program.

Parameter (units)	Method/Reference	Condition/ Holding Time	Method Detection Limits
Field (In situ)			
Temperature (^{o}C)	N.B.S. calibrated EPA 1979 #170	< 5 min.	0.1°C
Dissolved Oxygen (mg/L)	Membrane Probe EPA 1979 #360 Luminescent method ASTM-D888-09 (C)	< 5 min.	0.2 mg/L
pH	Glass Probe EPA 1979 #50	< 5 min.	0.1 units
Specific Conductance (umhos/cm)	Conductivity Bridge APHA #205	< 5 min.	% of calibration standard
Secchi Disc (cm) *estuarine stations*	20 cm Black/White	< 5 min.	0.1 meter
Laboratory			
Dissolved Organic Carbon (mg/L)	EPA Method 415.1	Frozen, 28 days/ 4 ^{o}C, 48 hrs	0.14 mg/L
Particulate Carbon (mg/L)	Exeter Analytical Model CE-440 Elemental analyzer	Frozen 28 days	0.006 mg/L
Ammonium (mg/L)	EPA Method 350.1	Frozen, 28 days/ 4 ^{o}C, 48 hrs.	0.004 mg/L
Particulate Nitrogen (mg/L)	Exeter Analytical Model CE-440 Elemental analyzer	Frozen 28 days	0.003 mg/L
Total Dissolved Nitrogen (mg/L)	alk. Persulfate then EPA 353.2	Frozen, 28 days/ 4 ^{o}C, 48 hrs.	0.034 mg/L
Nitrate + Nitrite (mg/L)	EPA Method 353.2	Frozen, 28 days/ 4 ^{o}C, 48 hrs.	0.003 mg/L
Nitrite (mg/L)	EPA Method 353.2	Frozen, 28 days/ 4 ^{o}C, 48 hrs.	0.002 mg/L
Orthophosphate (mg/L)	EPA Method 365.1	Frozen, 28 days/ 4 ^{o}C, 48 hrs.	0.002 μg/L
Particulate Phosphorus (mg/L)	Combustion, HCl extraction, then EPA Method 365.1	Frozen 28 days	0.003 mg/L
Total Dissolved Phosphorus (mg/L)	alk. Persulfate then EPA 365.1	Frozen, 28 days/ 4 ^{o}C, 48 hrs.	0.006 mg/L
Biochemical Oxygen Demand (BOD)	EPA Method 5210 B	4 ^{o}C 48 hrs.	NA
Total Suspended Solids (mg/L)	EPA Method 160.2	4 ^{o}C 7 days	1.88 mg/L
Total Dissolved Solids ppm	EPA Method 160.1	4 ^{o}C 7 days	2 ppm
Turbidity (NTU)	EPA Method 180.1	4 ^{o}C 48 hrs.	0.1 NTU
Chlorophyll “a” (μg/L)	Spectrophotometric SM 20th Ed. #10200 H	Frozen 28 days	0.62 μg/L
Phaeophytin “a” (μg/L)	Spectrophotometric SM 20th. Ed. #10200 H	Frozen 28 days	0.74 μg/L
Sulfate (mg/L)	EPA Method 375.4	4 ^{o}C 28 days	2 mg/L
Alkalinity, Total (mg/L)	EPA Method 310.1	4 ^{o}C 14 days	1 mg/L

MEASUREMENT/DATA ACQUISITION

B1 Program Design

Table 1 on pages 9-13 provides the station location descriptions for Maryland DNR's Ambient Water Quality Monitoring Program. Water quality data are collected at select stations in tributaries of the Choptank, Gunpowder, Patapsco, Patuxent, Susquehanna, Potomac and Youghiogheny Rivers. The selection of stations for this monitoring program was guided primarily by the need to assess conditions in water use areas. These included recreational areas, surface water supply areas, land use areas and potential areas of development. All ambient stations are sampled monthly (12 collections/year) on a pre-determined date. This sampling design allows the collection of data over a wide range of stream/river flows and provides adequate data for capturing long-term annual trends.

B2 Sampling Methods

Maryland DNR staff obtain field data and water quality samples (grab samples) from all stations by land (freshwater stations) or boat (8 estuarine stations). Bucket sampling is used on all land runs to collect samples from bridges, weirs, and stream banks. Bridge sampling is the preferred sample method and is used whenever possible. For the tidal stations, a submersible pump is used to collect a surface sample (at 0.5 m depth) and several other samples at various depths, from a boat. Appendix II provides the Standard Operating Procedures for Maryland DNR's Ambient Water Quality Monitoring Program (referred to as #PR-03: Maryland Core/Trend Monitoring Program).

B2.1 Field Measurements

Maryland DNR personnel obtain field data with the use of multi-parameter instruments. The operation and calibration protocols are outlined in Section 6.0 in MDNR's Standard Operating Procedures #PR-03: Maryland Core/Trend Monitoring Program (Appendix II). The instruments are maintained according to the operating manuals provided by the manufacturers for each instrument. Field measurements include dissolved oxygen, temperature, specific conductance and pH. These parameters are currently measured with instruments manufactured by Hydrolab or YSI. At the estuarine stations, secchi depth is measured with a 20 centimeter black and white secchi disc. Table 2 provides the methods and detection limits for the field measurements.

B2.2 Water Quality Samples

Grab samples for each station are collected by MDNR personnel utilizing methods dependent on the physical conditions of the station location. Estuarine stations (8) are sampled by boat, whereas the land run stations are sampled from a bridge, weir, or streamside. At each land run station, a surface grab sample is collected, if possible, at mid-channel. For a complete description of the collection methods utilized under each condition please refer to Section 8.0 in MDNR's Standard Operating Procedures #PR-03: Maryland Core/Trend Monitoring Program (Appendix II). Both whole water and filtered water samples for each freshwater station are provided to the Maryland Department of Health and Mental Hygiene's (DHMH), Environmental Chemistry Division for analysis. A complete list of the physical and analytical parameters obtained, holding times, methods, and method

detection limits is provided in Table 2 (page 15).

B3 Sample Handling and Custody

Laboratory samples are placed on ice (i.e., stored at 4°C) in a large cooler and transported by Maryland DNR field personnel to Annapolis. Iced samples are then shipped overnight directly to DHMH. Frozen samples are placed in a freezer in Annapolis and delivered weekly to DHMH. Table 2 provides the holding times for each water quality parameter. For a complete description of sample handling and the procedures utilized to label and track all samples, please see Sections 9 and 10 in MDNR's Standard Operating Procedures #PR-03: Maryland Core/Trend Monitoring Program (Appendix II). Because the data generated by MDNR's Ambient Water Quality Monitoring Program are not used for legal purposes, formal chain-of-custody procedures are not required.

B4 Analytical Methods

Table 2 provides a list of analytical methods for all water quality parameters utilized by MDNR's Ambient Water Quality Monitoring Program. All analysis (except chlorophyll "a" and phaeophytin "a") is conducted by the State of Maryland, DHMH Laboratories. The Standard Operating Procedures for all water quality parameters utilized by DHMH are detailed in Appendix III. For each water quality parameter, scope of application, methods, equipment and supplies, reagents and standards, sample collection, quality control, procedures, data analysis and calculations, and data management are detailed. Starting in January 2009, chlorophyll "a" and phaeophytin "a" is analyzed by the University of Maryland, Chesapeake Biological Laboratory, Nutrient Analytical Services Laboratory (NASL). These parameters were originally measured by DHMH. The analytical methods utilized by NASL are identical to the ones that were utilized by DHMH and are detailed in Appendix VIII of the Quality Assurance Project Plan for Maryland DNR's Chesapeake Bay Water Quality Monitoring Program – Chemical and Physical Properties Component. The latest version of this Plan is available at:
http://mddnr.chesapeakebay.net/eyesonthebay/documents/MdDNR_MTQAPP2015.pdf.

B5 Quality Control

The data collected as part of the Ambient Water Quality Monitoring Program are used in making management decisions regarding Chesapeake Bay water quality as described in section A5. DNR follows specific procedures to ensure that the design is properly implemented and that monitoring measurements are made and managed with sufficient accuracy, precision, and detection limits. Section 11.0 in Appendix II provides a list of quality control and quality assurance procedures that are utilized for this monitoring program. General discussions of quality assurance and quality control aspects associated with accuracy, precision, and audits are provided in the subsections below.

B5.1 Accuracy

The accuracy (closeness to the true value) of the collected data is controlled and assured by the proper use, calibration, and maintenance of both field and laboratory equipment for the measurement of physical and chemical parameters. All instruments are identified by a unique number, used as an index for documentation of calibration, repair, and preventive maintenance. Daily quality control checks (including the running of blanks and standards) are used to control and assure laboratory accuracy. Accuracy of laboratory results

is also assessed through DNR's participation in the Chesapeake Bay Coordinated Split Sample Program (CSSP), a split sampling program in which nine laboratories involved in Chesapeake Bay monitoring analyze the coordinated split samples.

B5.2 Precision

Precision of the chemical analytical methods is determined and documented from duplicate analyses. Every tenth sample is analyzed in duplicate at DHMH. Precision of the entire measurement system for laboratory-analyzed parameters, including field processing, storage, and chemical analysis, can be assessed from duplicate field samples. Duplicate field samples are routinely collected approximately every 10 to 20 samples.

B5.3 Audits

Performance audits for chemical analyses are based on the results of samples distributed by the EPA Chesapeake Bay Program Blind Audit Program. These samples must fall within the 95% confidence interval for acceptance. If results fall outside this range, corrective actions for each parameter and measurement are taken. The DNR Quality Assurance Officer communicates on a weekly basis with the field program staff and confers with the laboratory quality assurance officers to ensure that all aspects of the program are being conducted properly.

B6 Instrument/Equipment Testing, Inspection, and Maintenance

Field crews carry two calibrated multiparameter instruments in case of failure. The meter in use is compared to the reserve meter any time (a) the field scientist recording measurements observes values outside the "typically expected range"; (b) the meter generates variable or erratic values; or, (c) the meter in use displays an error message. If the meters do not agree within acceptable limits, the reserve meter is used. This is noted on the field data sheet (the field sheet is illustrated in MDNR's Standard Operating Procedures #PR-03: Maryland Core/Trend Monitoring Program (Appendix II)). All equipment repairs are handled by Greg Gruber, MDNR's Field Quality Assurance Officer for this monitoring program. All parts are ordered directly from the manufacturer. If the repairs cannot be performed by the Field Quality Assurance Officer, the instrument is sent to the manufacturer for repairs.

Laboratory instrument standard operating procedures include preventive maintenance procedures as well as performance checks and calibration procedures. Appropriate maintenance is scheduled based on the results of performance checks or after a specified number of hours of operation. Instrument preventive maintenance, repairs, and analytical corrective actions are documented on laboratory notebooks or on DHMH's Divisional Analytical Corrective Actions forms (Appendix IV).

B7 Instrument/Equipment Calibration and Frequency

As mentioned previously, Section 6.0 in MDNR's Standard Operating Procedures #PR-03: Maryland Core/Trend Monitoring Program provides detailed information regarding multiparameter instrument (Hydrolab or YSI) calibration. The following information provides general procedures that should be followed while utilizing multiparameter instruments for this monitoring program.

Calibration and Frequency

A. Set up a calibration logbook for each unit, with make, model, and serial number and purchase date. Assign a letter for MDNR use as required.

B. Calibrate meters on Friday for use the next week. After one to four days of field use, post calibrate equipment to determine if parameters have drifted.

C. Specific conductance calibration shall be made using standards generated by the field office from dry KCl and deionized water. Standards used are 294, 720, 2767, 6668, 12950, and 24820 microsiemens/cm (microsiemens=microS=μS); or 0.002, 0.005, 0.02, 0.05, 0.1, and 0.2 molar KCl, respectively. (At 25 °C microsiemens/cm = micromhos/cm.)

D. A pH calibration shall be made using premixed standards of color-coded pH 4.00, pH 7.00 and pH 10.00 purchased from Fisher Scientific. Standards are specifically labeled (contain expiration dates) and color coded - red for pH 4.00, yellow for pH 7.00 and blue for pH 10.00.

E. The dissolved oxygen calibration method is dependent on the type of dissolved oxygen probe used on the instrument. If the instrument is outfitted with a standard Clark cell, the dissolved oxygen calibration shall be done on the common standard of water-saturated air. After correcting for the barometric pressure and temperature, the oxygen content of water-saturated air can be checked against standard DO tables. The DO membrane is also visually checked every time the meter is pre- or post-calibrated. If the membrane appears damaged, the meter is posted as is. Then the membrane and electrolyte are replaced and the meter is calibrated after 24 hours. If the instrument is outfitted with an optical probe (LDO for Hydrolab; ROX for YSI) then the standard of air-saturated water is used. After correcting for the barometric pressure and temperature, the oxygen content of the air-saturated water can be checked against standard DO tables. The cap (LDO) or membrane (ROX) should also be visually checked every time the meter is pre- or post-calibrated.

F. Record all pre-calibration, post-calibration, and maintenance procedures in the log book, including any values (e.g. barometric pressure, temperature) that are used in the calibration procedures. An example of the equipment calibration log is included.

G. Record any unusual circumstances that may affect the instrument readings in the logbook.

B8 Inspection/Acceptance of Supplies and Consumables

From 1974 to September 2007, the deionized water used at the MDNR Annapolis field office was generated from Annapolis City water passed through a non-pressurized Barnstead cartridge system equipped with two Ultrapure mixed bed cartridges and one organic removal cartridge. This produced ASTM Type III water. Starting on September 10, 2007, the MDNR field office switched to a system that produces ASTM Type II water. The deionized water is generated from tap water using a Thermo Scientific Barnstead DIamond TII RO/DI system with a GE SmartWater external pre-filter. The RO/ DI system is linked to a Thermo Scientific Barnstead DIamond TII 60L storage reservoir. The system uses a thin film composite reverse osmosis membrane with pretreatment to produce RO water. This water is then put through a two-stage deionization process combined with UV oxidation and a 0.2 micron final filter. The reagent grade water provided by this system exceeds ASTM Type II and NCCLS/CAP Type I standards. All manufacturer recommendations are followed regarding cartridge replacement and system sanitation (Refer *Apr 17, 2008, Revision 14, QAPP: Chemical & Physical Property Component Page VI-3* to Thermo Scientific. 2007. Barnstead DIamond TII Type II Water System Operation Manual and Barnstead DIamond TII Type II Storage Reservoir Operation Manual). The GE SmartWater pre-filter was placed inline to improve the integrity of feed-water going into the Barnstead DIamond System. The pre-filter is changed at least every three (3) months or more frequently during periods of heavy use. A log is kept at the front of the DI System Manual to document all changes and updates made to the system.

The Maryland Department of Health and Mental Hygiene produces deionized water by utilizing a water system provided, serviced and set up by Millipore. In this system, tap water is passed through a 1 micron filter, a carbon tank, and two mix bed ion exchange resin columns. The water is then subjected to UV oxidation and passed through a 0.2 micron filter. Appendix III (DHMH's Standard Operating Procedures for Water Quality Parameters) lists all supplies and consumables utilized by the analytical laboratory for sample analysis.

B9 Non-direct Measurements

No data are needed for implementation or decision-making that are obtained from non-direct measurement sources.

B10 Data Management

Data collection for the Ambient Water Quality Monitoring Program begins when measurements from field recording instruments are entered onto field data sheets. A section on the field sheet is used to document any problems encountered in the field that might affect the field parameters or the samples brought back for the laboratory analysis. A senior field scientist ensures that all measurements are taken and recorded properly. After field personnel have completed data sheets for a given calendar month, they make a photocopy of the sheets to keep in the Field Office, and send the original field sheets to data management staff at the MDNR Tawes State Office Building in Annapolis. The Field Office also generates a Cross Reference Sheet for each set of field sheets, which is sent to the DNR data management personnel along with the field data sheets. The Cross Reference Sheet provides the data management personnel with the documentation to determine what field and laboratory results to expect. The Cross Reference Sheet is illustrated in Appendix B contained in MDNR's Standard Operating Procedures #PR-03: Maryland Core/Trend

Monitoring Program (Appendix II).

Laboratory analysis sheets are also initiated in the field (laboratory sheet is illustrated in Appendix D MDNR's Standard Operating Procedures #PR-03: Maryland Core/Trend Monitoring Program (Appendix II)). These laboratory sheets list each parameter requested for analysis and include basic information about the sample, such as station, date, time, and depth. The sheets serve as sample transfer sheets, traveling with the samples to the Maryland Department of Health and Mental Hygiene laboratory (DHMH) for analysis. Both the sheets and the samples are logged in at the laboratory.

The laboratory analyst reviews the data and, if the data exceed their control limits, the entire run is re-analyzed. Re-analysis can occur for any number of reasons, such as, a poor r-squared on the standard curve, the wrong set of pump tubes (which would provide abnormally low peaks), or high blank values (in the case of DOC). Once laboratory staff has completed the laboratory sheets, they are sent to the DNR data management at the Tawes Building.

Data review and verification are conducted at four levels by DNR data management personnel. At the first level, DNR data management personnel review cross reference sheets and field data sheets: (1) comparing field sheets to cross reference sheets to ensure that all field sheets have been received; (2) reviewing all field sheets to check that they are filled out completely and legibly, and; (3) sending the sheets to a data entry service for keypunch. At the data entry service, the field sheet data are double-entered to minimize errors at the keypunch stage. The entered field data are sent back to DNR as an electronic file for further processing.

At the second level, a Data Processing Technician generates reports and plots for data verification using the Water Quality Import v3 software. The WQ Import v3 software was designed in late 1998 and completely developed in 2000 in Microsoft Access. The WQ Import v3 software is used to conduct data management activities, such as performing an initial data check, conducting major key field checks, performing a parameter range check (including measured and calculated parameters), conducting combination checks for specific parameters, generating an error report and verification plots, generating a "data verified list," reforming data, creating a database, and submitting data.

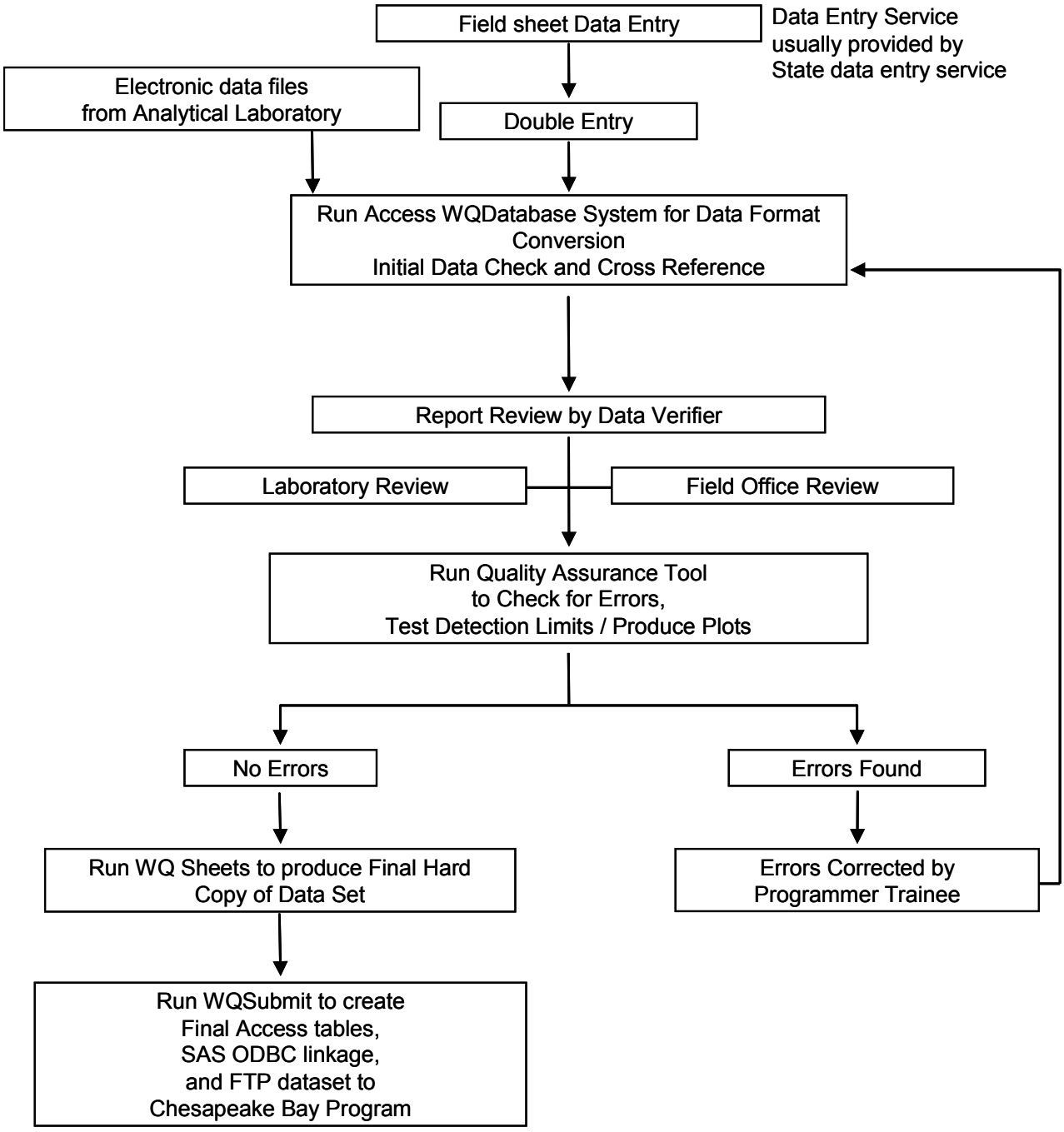
Third, system printouts or PDF files of each data set are sent to a biologist and the Quality Assurance Officer for verification and editing. The Quality Assurance Officer and DNR biologists ensure that measured or calculated information for all types of data are correct and that the codes associated with parameters are properly established. In addition, the Quality Assurance Officer identifies data problems, provides data correction instructions, and coordinates data correction activities. Possible errors are identified, and sent to the laboratory or field office for verification or verified over the phone. Any necessary corrections are written on an edit form, which is given to a Data Processing Technician. The technician makes changes to correct the electronic data set, re-runs the verification programs, and updates the verification reports and plots. This procedure is repeated until a clean data set is produced.

The fourth step is for data management staff to ensure that the overall data verification processes are completed, all data errors are corrected, and that the finalized data sets are created and formatted to be consistent with historical data sets.

The final data set combining the field and laboratory data is created as an Access "MDB file" after the completion of data verification processes. This final data set is stored in a local designated DNR database directory for data user access. Data requests should be directed to Mark Trice, Program Chief of Water Quality Informatics (410-260-8630). A formatted submission data set and associated data documentation is also transferred to the Chesapeake Bay Program Data Center on a monthly basis.

Files submitted by DNR are further screened by the Bay Program's Quality Assurance Tool (QAT). The QAT generates a Chesapeake Information Management System (CIMS) Water Quality Data Quality Assurance Report for each file submitted. The Report identifies fatal errors for records that are incompatible with the CIMS database and prevents files that have fatal errors from being entered into CIMS. Nonfatal errors, such as those with values falling out of a historical range are reviewed and accepted or rejected. Once data are entered into the CIMS database, they are available to the public via the Bay Program datahub at: www.chesapeakebay.net. The data management process is diagramed in Figure 3.

Figure 3. Data Management Flow Chart: Data Entry through Production of Final Master Data Set



ASSESSMENT AND OVERSIGHT

Program and performance audits verify that procedures specified in the Project Plan are being followed throughout the entire MDNR Ambient Water Quality Monitoring Program. These audits ensure the integrity of the reported data so they can be used for their intended purpose.

C1 Assessments and Response Action

C1.1 Field Activities

If a station or specific sample cannot be collected, it is noted on the cross-reference sheet. Specific problems associated with field collection of a site are also noted on the field sheet. Conditions that may affect data results are included in the comments section of the field sheet so that they are available to each data analyst. If post calibration results are outside acceptable limits, the individual calibrating notifies the Field QA Officer who decides if data should be deleted or flagged. If split sample results suggest that there is a problem with the data, the issue is thoroughly discussed by laboratory and field representatives at DIWG and possible solutions are offered. The Field QA Officer regularly reviews Equipment Log Books to ensure that all staff is following QC procedures. Standard maintenance recommended by Hydrolab and YSI is performed at twelve-week intervals. All serious Quality Control issues are reported directly to the Field Office Project Chief. Audits of the field staff are conducted as needed.

C1.2 Laboratory Activities

Corrective actions are initiated by the analyst, with the input of the Lead Scientist of the Laboratory Section, if necessary. The Lead Scientist and the Supervisor review corrective actions. A copy of the completed form is submitted to the division QA officer, and the original is kept in the laboratory. The DHMH Division of Environmental Chemistry is audited approximately every three years by EPA Region III or Chesapeake Bay Program Office staff.

C1.3 Data Management Activities

The Data Input Editor is the first line of defense for data correction. Maryland DNR data management personnel review all incoming data and compare the data to the cross-reference file. Data management personnel verify the submitted data and apply corrections to the physical datasheet if errors are identified. During the data-import process, a Data Processing Technician makes all corrections to the data and key fields as they are imported into the WQ Database System. The Data Processing Technician assists where needed in constructing better tools to edit and apply to large quantities of data corrections if necessary. Documenting the correction is handled within WQ Maintenance process. If the correction is fairly generic, edits to the changes are logged. There is no formal documentation for editing data sheets. These tasks are considered extreme and performed only when confirmed by field office or laboratory personnel.

C2 Reports to Management

Maryland DNR provides four quarterly progress reports (January, April, July and November) that indicate the number of Ambient Water Quality Stations that were sampled and an update on the status of water clarity assessments, efforts to acquire and assess data for the Integrated Report and a copy of the State 305(b) update. Status reports also provide explanations, if needed, for why accomplishments fell short of the projections. In addition, any changes to the Quality Assurance Project Plan or the SOPs referenced herein are documented and approval is required for all of those involved in project management (i.e., the individuals responsible for the major aspects of this monitoring program).

DATA REVIEW AND USABILITY

D1 Data Review, Verification, and Validation

Field: Described in C1.1 above.

Laboratory: The DHMH Environmental Chemistry Division uses data review checklists for data validation (example provided in Figure 4). Appendix III provides copies of each data review checklist for all water quality parameters.

Data Management: The data-management group validates key fields. The key fields ensure that the data are accurate and will not be lost or duplicated within the system. After the key fields are validated, the values are plotted to discover any anomalies. The scientists/project managers review the reports and determine if any additional edits are needed to data values. These edits are reported to the Data Processing Technician who makes the changes (Figure 3).

D2 Verification Validation Methods

Maryland DNR data management personnel conduct data review and verification at four levels: At the first level, DNR data management personnel review cross reference sheets and field data sheets: (1) comparing field sheets to cross reference sheets to ensure that all field sheets have been received; (2) reviewing all field sheets to check that they are filled out completely and legibly, and; (3) sending the sheets to a data entry service for keypunch. At the data entry service, the field sheet data are double-entered to minimize errors at the keypunch stage. The entered field data are sent back to DNR as an electronic file on a diskette for further processing.

At the second level, a Data Processing Technician generates reports and plots for data verification using the Water Quality Import v3 software.

At the third level, system printouts of each data set are sent to a biologist and the Quality Assurance Officer for verification and editing. The Quality Assurance Officer and DNR biologists ensure that measured or calculated information for all types of data are correct and that the codes associated with parameters are properly established.

At the fourth level, data management staff ensure that the overall data verification processes are completed, all data errors are corrected, and that the finalized data sets are created and are formatted to be consistent with historical data sets. The final data set combining the field and laboratory data is created as an Access database file after completion of data verification processes. This final data set is stored in a local designated DNR database directory for data user access. A formatted submission data set and associated data documentation is also transferred to the Chesapeake Bay Program Data Center on a monthly basis.

D3 Reconciliation with User Requirements

The data generated by Maryland DNR's Ambient Water Quality Monitoring Program are utilized to calculate relative status and long-term linear and non-linear trends (Appendix I describes methods). These calculations are performed for MDNR under contract by a statistical consultant. The experimental design of this program requires monthly collections of water quality data (i.e., 12

collections per year) which are adequate for capturing long-term annual trends (Alden et al., 1994). As a result, the data generated by this program directly meet the objectives for which it is collected.

Figure 4. Data Review Checklist
DIVISION OF ENVIRONMENTAL CHEMISTRY
INORGANICS ANALYTICAL LABORATORY

Data Review Checklist LL Orthophosphate/ LL Ammonia

EPA Method 365.1/ EPA Method 350.1

Lab Numbers: [1] _____

Analyst: _____

Dates Collected: _____ **Date Analyzed:** _____

Procedure	Acceptance Criteria	Status(√)	Comments
Holding Time	48 hours @ 4°C; 28 days @ –20°C		
Calibration Curve	Corr. Coeff. ≥ 0.9950		
Reagent Blank	< Reporting Level (0.004 ppm for OP; 0.008 ppm for NH_3)		
Blank Spike	1 per batch		
	Recovery = 90–110%		
Matrix Spike	Every 10th sample or 1/batch, if less than 10		
	Recovery = 90–110%		
External QC	Beginning and end of each run		
	Within acceptable range		
Check Standard	After every 10th sample and at the end of the		
	Concentration = 90–110% of the true value		
Duplicates/Replicates	Every 10th sample or 1/batch, if less than 10		
	RPD ≤ 10%		
Decimal Places Reported	3		
Measured Values	Within calibration range (0.004–0.250 ppm for OP; 0.008–0.500 ppm for NH_3)		
Diluted Samples	Correct final calculations		
Changes/Notes	Clearly stated		

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Appendix I: Methods for Calculating Status and Trends at Maryland DNR's Ambient Water Quality Monitoring Stations

Status and trend analyses are performed at all CORE/Trend monitoring stations for nutrients (nitrogen and phosphorus), field-measured parameters such as conductivity, pH, and water temperature, and total suspended solids, total alkalinity, and total organic carbon. Trends in chlorophyll and sulfate are also assessed at stations where those parameters are measured.

Water quality status is a measure of the current water quality condition compared to some benchmark. Trends are measured to assess changes in water quality over time. For the CORE/Trend stations increasing trends are generally associated with degrading water quality and decreasing trends are generally associated with improving water quality conditions. Linear and non-linear trend analyses are performed on "long-term" data (1986 to present) and "short-term" data (1995 to present). Trends for both long- and short-term periods are deemed significant if the probability value (p-value) is less than or equal to 0.01, a level which was chosen to reduce the Type I error rate (i.e., reduce the chance of stating that there is a trend when in fact there is no trend).

Relative Status

In the absence of water quality reference levels or restoration targets, members of the Data Analysis Workgroup (DAWG), which was a workgroup of the U.S. Environmental Protection Agency Chesapeake Bay Program Monitoring Subcommittee, developed a method of comparing stations in similar salinity zones (Aden and Perry, 1997). The first version of the relative status method was developed and implemented for the 1997 Re-evaluation effort of the Chesapeake Bay Program to assess progress in meeting nutrient reduction goals established in 1987.

The relative status method compares the median of the most recent three years of data for a specific parameter at a particular station to all stations in the same salinity regime using "cut-points" in a benchmark data set. The most recent three years of data are used in the status data set to even out year-to-year climatic variation. The benchmark data set consists of water quality data that were collected between January 1985 and December 1990 to establish a Bay-wide baseline against which to score water quality at each station. Data in the benchmark data set were partitioned using a beta cumulative distribution function to establish the "cut-points" for individual parameters in a particular salinity zone; in this case non-tidal, fresh water systems.

The status data set is scored using the logistic probability integral transform, and the score is adjusted based on sample size to account for the inter-dependence of observations. The lack of independence in observations at a site tends to result in too many observations in the ends of the distribution, i.e., in the "good" and "poor" categories. The adjustment results in a more even distribution of scores. Both the benchmark and status data sets are log transformed prior to analysis to meet the distributional assumptions of the procedure.

It is important to note that this is a relative ranking procedure. Just because a parameter at a station is scored as "good" does not mean that water quality is acceptable. It is only a relative indication that the station scored better than other stations rated as "fair" or "poor" based on a comparison to cut-points in the benchmark data set for similar stations.

Linear Trend Analysis

A combination of statistical procedures is used to calculate the significance of linear trends and the percent change in a water quality parameter over time. First, the seasonal Kendall test, which is a generalization of the Mann-Kendall test, is used to assess the presence of a trend in time in seasonal data (Hirsh, et al., 1982). The seasonal Kendall test was proposed by Hirsh for use in

data with 12 seasons (months). Basically, the Mann-Kendall "S" statistic and its variance VAR(S) are computed for each month (season) for data collected over a number of years. The seasonal statistics are then summed over the years and an overall Z statistic is then calculated, which is compared to standard normal tables to assess significance (Gilbert, 1987). The seasonal Kendall test is particularly useful for water quality trend assessment because it is non-parametric (i.e., does not assume any particular distribution in the data), and is not affected by missing, tied, or below detection limit values.

The magnitude of change in the data over time, for trends that are significant, is calculated using the seasonal Kendall slope estimator, which is a generalization of Sen's estimator of slope (Sen, 1968). The seasonal Kendall slope estimator is calculated from the median of all possible differences in the data. The magnitude of change is expressed as the percent change since the beginning of the period of record. Percent change is calculated by multiplying the Sen's slope estimator times the number of years of the study period, and dividing by the initial median:

$$\text{Percent change} = [(\text{slope}*\text{nyrs})/\text{base median}] * 100$$

The base median is calculated from the first two years of data. For most stations that period is January 1986 to December 1987.

Non-linear Trend Analysis

As the monitoring timeline has lengthened, non-linear trend analyses have been added. The utility of identifying non-linear trends was investigated by Alden (Alden et al., 2000). Non-linear trends provide a picture of how a parameter has changed within the period of evaluation and can serve as early warning signals for managers. These analyses identify whether a trend is primarily linear (generally unidirectional), U-shaped (decreasing early in the time series, increasing later in the time series), or the reverse (inverse U-shaped). They can also determine where the critical point (change in direction of trend), if any, is located (either within or outside the period of evaluation).

Non-linear trends are assessed using a general linear model with a linear TIME term and a quadratic ($TIME^2$) term. Data are log-transformed prior to the analysis in an effort to conform to the distributional requirements of the general linear model. A MONTH term is included in the model to account for seasonal effects. The TIME and $TIME^2$ terms are first standardized to a mean of zero to ensure that the TIME and $TIME^2$ terms are orthogonal (uncorrelated), thus reducing the chances of misinterpretation of correlated regression terms (E. Perry, personal communication).

The log of the water quality parameter in question is then regressed on TIME and $TIME^2$ using SAS® PROC GLM (SAS, 1989) with the following statements:

```
PROC GLM;
  CLASS MONTH;
    MODEL log(WQ variable) = TIME TIME2 MONTH;
```

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Appendix II: Maryland Department of Natural Resources: Standard Operating Procedures #PR-03: Maryland Core/Trend Monitoring Program

Standard Operating Procedure # PR-03
MARYLAND CORE/TREND MONITORING PROGRAM
Revision 2: March 2015

Prepared by: Kristen Heyer

Reviewed by: Laura Fabian

Approved by: Sara Bowen

Table of Contents

1.0 Scope and Application

1.1 This Standard Operating Procedure is applicable to the collection of water quality samples for the Maryland Core/ Trend Program (Section 106).

1.2 The water samples are collected for physical and chemical analysis.

2.0 Summary of Method

2.1 Stations sampled for the Core/ Trend Monitoring Program are mostly sampled by land. A few Core/Trend stations are sampled in conjunction with other programs: Chesapeake Bay Mainstem (refer to SOP# PR-01); Chesapeake Bay Tributary (refer to SOP# PR-02); and Potomac River Program (refer to SOP# PR-05). These samples are collected at depth, aboard a research vessel and follow the respective program Standard Operating Procedure.

2.2 Whole water samples are collected at every station for the Core/ Trend Program. The following equipment are used for collection of water samples depending on the station description and conditions:

- Bucket
- Submersible pump

2.3 The whole water samples are collected and analyzed for both physical and chemical properties; whole water nutrients and dissolved fractions.

3.0 Health and Safety Warnings

3.1 When sampling from boats and piers, wear appropriate safety gear and follow appropriate safety procedures for working around the water and under slippery conditions.

3.2 When sampling from road bridges, wear appropriate safety gear and follow appropriate safety procedures for working around high speed traffic.

3.3 When sampling along the stream bank, care should be exercised in areas with slippery and uneven terrain.

3.4 Proper methods for lifting and moving equipment and samples should be exercised to avoid bodily injury.

3.5 Any water sample collected could contain a potentially harmful algae species. If the presence of a toxic species is suspected, protective equipment **must** be used. Protective gear must include gloves and raingear. Respirators and goggles must be used when sampling a bloom suspected of containing algae species that can produce toxic aerosols such as *Pfiesteria sp.*

4.0 Interferences

4.1 Contamination of samples can be minimized or eliminated by following the procedure for cleaning of sampling equipment. Equipment is washed on a regular basis to include acid rinsing. Refer to SOP # MC-01: *Cleaning and decontamination of sampling equipment*.

4.2 Improper sample collection can be avoided by following the guidelines in this SOP and taking care not to disturb the substrate of the area being sampling.

4.3 Care should be exercised to avoid collecting debris in the sample when it is present in the body of water being sampled.

4.4 Inaccessibility to sampling stations may occur due road and/or bridge construction, maintenance or closure. Inaccessibility can also be caused by boat ramp closure, homeowner denial of access to the site and overgrowth of brush.

4.5 Weather conditions may interfere with the collection of samples. High winds and ice cover are examples of interferences that may occur. Sample collection should be rescheduled if possible.

4.6 Insufficient water volume would interfere with the collection of grab samples. This problem could manifest itself as an inaccessibility problem (i.e. there is not enough water to reach the station) or there is not sufficient volume to grab a sample.

4.7 Malfunctioning sampling equipment, i.e. submersible pumps, etc. will interfere with the collection of grab samples. Malfunctioning or missing filtration equipment, i.e. electric vacuum pumps or filter funnels, will interfere with the generation of particulate and dissolved samples. Having spare filtration equipment and/ or a hand pump could solve this problem. If too many samples need to be processed by hand, the whole water sample used for filtering can be preserved in ice and filtered as soon as you return to the office. Be sure to note the time the sample was filtered on the volume sheet.

4.8 Contamination can occur from dirt and debris near the processing area. This can be minimized by maintaining clean vehicles, vessels and lab areas. Filtration equipment is placed on clean lab towels during processing to further maintain a clean working environment. The boat and/ or vehicle engines should not be running while processing samples if the fumes could accumulate in the processing area.

4.9 Insufficient lab supplies would also interfere with collection of particulate and dissolved nutrient samples. This problem can be solved by maintaining a back-up supply of necessary items.

5.0 Equipment and Supplies

5.1 A bucket is typically the equipment used for collection of water samples for the Core/ Trend Program. For bucket sampling, we use a square 3 gallon bucket (Rubbermaid, model #2964). The bucket is then tied to a line long enough to reach the water surface. The bucket should be cleaned periodically (refer to SOP # MC-01: *Cleaning and decontamination of sampling equipment*). Each van is equipped with its own sampling buckets. Bucket sampling will be described in this SOP.

5.2 A submersible pump is used for Core/ Trend stations that are sampled in conjunction with another program, i.e. Chesapeake Mainstem or Potomac. Submersible pump sampling uses either a well pump (Dayton, ½ HP, 230 V, model # D10KS05221) or a bilge pump (Rule, 2000 gph, 12 V, model # 10). Refer to the Standard Operating Procedure for the Field Collection of Grab Water Samples (SOP # SC-01) for sample collection details.

5.3 The whole water samples collected for the Water Quality Monitoring Program are collected in new HDPE (plastic milk jug) bottles. These may include, but are not limited to, half- gallon (2 qt. bottles), quart, 16 oz. and 8 oz. bottles. Ice-filled coolers labeled for courier delivery to the Baltimore Lab are necessary for transporting the whole water samples.

5.4 Any or all of the following equipment is used to collect and record data on the field sheets:

- pencils, pens & sharpies
- watch, clock or instrument that displays the current time
- thermometer (readings in Celsius)
- water quality instrument (Hydrolab/ YSI) with probe guard
- calculator (for boat stations sampled where there is a pycnocline)
- compass & secchi disk (for tidal stations)

5.5 Any or all of the following equipment is used for collection of particulate and dissolved samples.

- 25mm filter funnel, 200ml; polysulfone (Pall Corp.# 4203)
- 47mm filter funnel, 300 ml; magnetic (Pall Corp.# 4242)
- Filter funnel manifold; polyurethane (Pall Corp.# 4205) with trap
- 47mm filter funnel & base; Millipore
- Filter flasks, 1000ml/ 500 ml (Fisher# 10-181F; 10-180E)
- Graduated Cylinders; 10ml, 50ml, 100ml, 250ml
- Forceps
- DI squirt bottles

- Adjustable vacuum pump (115V AC or 12 V DC Air Cadet) with pressure gage and trap

5.6 Any or all of the following supplies are used for collection of particulate and dissolved samples. When sampling Core/ Trend stations that are sampled in conjunction with another program, i.e. Chesapeake Mainstem or Potomac additional supplies may also be used. Refer to the Standard Operating Procedure for the Field Filtration for Particulate and Dissolved Nutrient Constituents (SOP # SC-03) for full filtration details for other programs within the Water Quality Monitoring Program.

- Pads
  - CHLA & PP: 47mm GF/F Whatman glass fiber filter (#1825-047, Fisher # 09-874-71); pore size 0.7 µm.
  - PC/ PN: 25mm GF/F Whatman glass fiber filter, pre-combusted at 490^{o}C; pore size 0.7 µm. Direct from lab.
- 8 oz. or 16 oz. HDPE bottles
- DI water
- Sample water
- Freezer or cooler with ice for sample storage and/ or transport

6.0 Instrument Calibration

6.1 Hydrolab Series 4a Calibration

1. Temperature – temperature is measured with a stainless steel thermistor. Calibration is not required because it is factory set and not user adjustable. During periodic routine servicing, temperature is checked at one temperature against a standard mercury thermometer. The multiprobe is returned to the manufacturer if test results are not within 0.2°C of standard thermometer.

2. Dissolved Oxygen – dissolved oxygen is measured with a Standard Clark Polarographic cell and corrected to standard temperature and pressure and for specific conductance. The probe is calibrated using a 1 point mg/L linear protocol in water saturated air. Calibration dissolved oxygen concentration is determined from a table of saturation concentrations in mg/L as calculated from the instrument temperature and local barometric pressure measured with a Standard Fortin Mercury Barometer.

3. Specific Conductance – conductivity is measured with a probe having two opposing graphite electrodes oriented horizontally inside a vertical plastic channel. The conductivity reading is corrected to standard temperature (25°C). The probe is calibrated with a standard potassium chloride solution using a 2 point linear protocol. These standard solutions are made in house. The zero point is calibrated in air with the probe dry. The slope is calibrated

with a standard potassium chloride solution. The slope standard is selected so that field measurements fall between zero and the specific conductance of this standard, but as close to the specific conductance of this standard as possible.

4. pH – pH is measured with a two probe system (*in situ* pH and reference probes) and corrected to standard temperature. The *in situ* pH probe is a standard silver/silver chloride glass probe and the reference probe is a pellet of silver inside a hollow plastic sleeve containing 4 M potassium chloride solution saturated with silver chloride. This sleeve has a porous Teflon™ junction at one end to connect this probe to the environment. The pH system is calibrated with standard pH buffers using a two point linear protocol. The zero point is first calibrated with a pH 7.00 standard buffer. The slope is calibrated with either pH 4.00 or 10.00 standard buffer. The slope buffer is selected depending on pH measurements anticipated during field deployment.

6.2 Hydrolab Series 5 Calibration

1. Temperature – temperature is measured with a stainless steel thermistor. Calibration is not required because it is factory set and not user adjustable. During periodic routine servicing, temperature is checked at one temperature against a standard mercury thermometer. The multiprobe is returned to the manufacturer if test results are not within 0.2°C of standard thermometer.

2. Dissolved Oxygen – dissolved oxygen is measured with a Standard Clark Polarographic cell and corrected to standard temperature and pressure and for specific conductance. The probe is calibrated using a 1 point mg/L linear protocol in water saturated air. Calibration dissolved oxygen concentration is determined from a table of saturation concentrations in mg/L as calculated from the instrument temperature and local barometric pressure measured with a Standard Fortin Mercury Barometer.

3. Specific Conductance – conductivity is measured with a probe having two opposing graphite electrodes oriented horizontally inside a vertical plastic channel. The conductivity reading is corrected to standard temperature (25°C). The probe is calibrated with a standard potassium chloride solution using a 2 point linear protocol. These standard solutions are made in house. The zero point is calibrated in air with the probe dry. The slope is calibrated with a standard potassium chloride solution. The slope standard is selected so that field measurements fall between zero and the specific conductance of this standard, but as close to the specific conductance of this standard as possible.

4. pH – pH is measured with a two probe system (*in situ* pH and reference probes) and corrected to standard temperature. The *in situ* pH probe is a standard silver/silver chloride glass probe and the reference probe is a pellet of silver inside a hollow plastic sleeve containing 4 M potassium chloride solution saturated with silver chloride. This sleeve has a porous Teflon™ junction at one end to connect this probe to the environment. The pH system is calibrated with standard pH buffers using a two point linear protocol. The zero point is first calibrated with a pH 7.00 standard buffer. The slope is calibrated with either pH 4.00 or 10.00 standard buffer. The slope buffer is selected depending on pH measurements anticipated during field deployment.

6.3 YSI 6820/ YSI 6920 Calibration

1. Temperature – temperature is measured with a sintered metallic oxide thermistor. Calibration is not required because it is factory set and not user adjustable. During periodic routine servicing, temperature is checked at one temperature against a standard mercury thermometer. The multiprobe is returned to the manufacturer if test results are not within 0.2°C of standard thermometer.

2. Dissolved Oxygen – dissolved oxygen is measured with a ROX optical (luminescent) probe. The probe is calibrated using a 1 point percent saturation linear protocol in air saturated water. Calibration dissolved oxygen concentration is determined from a table of saturation concentrations in mg/L as calculated from the instrument temperature and local barometric pressure measured with a Standard Fortin Mercury Barometer.

3. Specific Conductance – conductivity is measured with a probe with four nickel electrodes. The conductivity reading is done as close to standard temperature (25°C) as possible. The probe is calibrated with a standard potassium chloride solution using a 1 point linear protocol. These standard solutions are made in house. The zero point is factory calibrated and cannot be calibrated. The slope is calibrated with a standard potassium chloride solution. The slope standard is selected so that field measurements fall between zero and the specific conductance of this standard, but as close to the specific conductance of this standard as possible.

4. pH – pH is measured with a bulb-type combination probe and is corrected to standard temperature. The pH probe consists of a proton selective glass reservoir filled with buffer (approx. pH 7) and a silver/silver chloride reference electrode that utilizes gelled electrolyte.

The pH system is calibrated with standard pH buffers using a two point linear protocol. The zero point is first calibrated with a pH 7.00 standard buffer. The slope is calibrated with either pH 4.00 or 10.00 standard buffer. The slope buffer is selected depending on pH measurements anticipated during field deployment.

6.4 Frequency of Calibration

- All instruments are calibrated prior to field use. Calibration is typically completed the Friday before the sampling week. If office time is available on a closer day preceding the run, then the calibration will be completed at that time.
- If large adjustments to the DO value were necessary during calibration, the DO should be checked prior to taking the instrument in the field.
- The DO is checked in the morning prior to sampling, when large changes in barometric pressure have occurred. This is typical of the Western Maryland Core run.
- Instrument calibrations may be rechecked in the calibration lab any time the field readings seem suspect.
- Instruments are post-calibrated (checked) after use. Typically the post-calibration occurs on the Friday after the sampling week. After post-calibration, the instrument is ready to be calibrated for field sampling the following week.

7.0 Preparation for Sampling

1. When preparing for a Core/ Trend sampling run the first step is to get the field pack for the specific run you are doing (prepped folders are kept in the common prep area in the back garage). The field packs contain all information to complete the run: field sheets, chemistry sheets, volume sheets, directions to all of the stations with maps, list for samples being collected, foils squares, baggies, PC/PN pads, lab towels, and extra sheets. When the run is complete, everything (except for the volume & chemistry sheets which accompany the samples) should be returned to the field pack and turned in to Laura Fabian.
2. Ensure that the field van has necessary supplies for safe collection of the sample. Each van should have buckets with line, orange cones and safety vests.
3. Ensure that the field van has the necessary supplies for processing the sample. Make sure there are enough quart, two quart and 16 oz. bottles for the whole and filtered samples. Check for a WORKING vacuum pump and appropriate filtration supplies: forceps, $MgCO_3$, Whatman pads.
4. Just prior to leaving for the run, the following equipment should be loaded into the van: fully charged & calibrated Hydrolab meters (294 µs/cm for specific conductance); filter unit, courier cooler big enough for all of the sample bottles, another small cooler for bringing pads back to the office after delivery to the courier and ice.

5. If your instrument has a Standard Clark cell, turn it on before arriving at the station so that it can warm up for 10 minutes before recording the readings.

8.0 Sample Collection

8.1 Bucket Sampling

Bucket sampling is used on all land runs, including Core/ Trend sampling. Bucket samples are taken from bridges, weirs and stream banks. However, bridge sampling is the preferred sample method and should be used whenever possible. Below, the steps to be taken for each type of bucket sampling are listed.

8.1.1 Bridge Sampling

NOTE: When sampling from bridges, always make sure you have the appropriate lengths of rope before leaving for your run. Once you arrive at the station:

1. Select the appropriate length of rope for the bridge from which you will be sampling. You may need to tie 2 or more ropes together to reach the water surface at some stations. Secure the rope to the bucket, making sure that it will not come loose while retrieving the sample. Most sample buckets already have the ropes tied on to ensure that the bucket "hangs" properly.
2. Sample on the upstream side of the bridge (if possible) and as close to the center of the stream/river as possible, where the majority of flow is located.
3. Lower the bucket to the water.
4. Tip the bucket and fill with enough water to rinse the bucket (at least a few inches). Depending on the height of the bridge, you may want to shake the rope to expel the rinse water from the bucket, or pull the bucket back up to dump the rinse water out of the bucket. Follow this procedure 3 times; making sure the bucket is properly rinsed.
5. Fill the bucket as full as possible.
6. Pull the bucket back up, making sure the rope does not rub against the side of the bridge. This can sometimes cause dirt, rust, paint, etc. to fall into the sample.
7. Carry the bucket back to the van.

8.1.2 Weir Sampling

NOTE: You may want to wear waterproof boots or water shoes when sampling from weirs or stream banks. Depending on the depth of the water, you may need to walk into the stream to collect a sample.

1. Select the shortest length of rope, and secure to the bucket.

2. Depending on the water level, you may be able to walk out onto the weir to get closer to the center of the stream. If the water level is too high, sample from the stream bank close the weir.
3. Always sample just upstream of the weir. Holding onto the rope, throw or drop the bucket into the water and fill with enough water to rinse without disturbing or touching the bottom of the stream with the bucket.
4. Dump the rinse water downstream of the sampling area.
5. Repeat steps 3 & 4 two more times.
6. Once the bucket is properly rinsed, throw or drop the bucket into the water and fill to the lip of the bucket.
7. Pull the bucket from the water, and carry it back to the van.

8.1.3 Stream Bank Sampling

1. Select the shortest length of rope, and secure to the bucket..
2. Try to get as close to the center of the stream as possible when it is safe to do so (using rocks, etc.). If it is shallow near the sample area, do your best to get a sample without touching or disturbing the bottom of the stream with the bucket. At times, you may have to walk out farther into the stream.
3. Holding the rope, throw or drop the bucket into the water and fill with enough water to rinse.
4. Dump the rinse water downstream of the sampling area.
5. Repeat steps 3 & 4 two more times.
6. Once the bucket is properly rinsed, throw or drop the bucket into the water and fill to the lip of the bucket.
7. Pull the bucket from the water, and carry the sample back to the van.

8.2 Recording the physical data

After the bucket has been collected, the next step is to take readings of physical parameters and record them on the field sheet.

8.2.1 Completing the Field Sheet

For complete details, see Standard Operating Procedure # DR-01: *Recording Physical Data on Field Sheets*. For an example Core/ Trend field sheet, see Appendix IV.

1. The start date is recorded as YYMMDD (2 digit year, 2 digit month, 2 digit day). Do not record an end date, it is assumed that it is a one day sampling period.
2. The start and end times are recorded in military time (4 digits). The start time is when you start sampling the station (collecting the bucket) and the end time is when you finish with the readings and whole water sample collection.

3. The two digit number of samples reflects the number of water samples collected at individual, discrete depths. It also denotes if there was a duplicate sample taken.
4. The total depth is only recorded on tidal stations. This is the depth, in meters, to the bottom.
5. Air temperature is recorded in Celsius to the nearest 0.5 degree. The thermometer should be hung out when you arrive on station to allow it to equilibrate to the current temperature. The thermometer should be placed approximately 3-4 feet from the ground and in the shade. Hanging the thermometer on the van in times of weather extremes may skew the reading.
6. Weather codes recorded on the field sheet are as follows:

    10- no precipitation 11- drizzle
    12- rain 13-heavy rain
    14- squally 15- frozen precipitation

    Yesterday's weather code is recorded as the predominate weather the day prior to sampling for the station being sampled. Today's weather code is the weather conditions at or near the time of sampling. If a weather condition had occurred that will affect the water quality sample, but does not represent today or yesterday's weather it can be noted in the comment section of the field sheet: e.g. 2 feet of snow covering the ground or hurricane 2 days ago.
7. Percent cloud cover is reported as a value from 0% to 100%. Numbers are three digits and are right justified on the field sheet: e.g. 005 would be 5% cloud cover. Thin clouds and haziness may be noted in the comment section of the field sheet.
8. Wind direction & velocity, wave height, secchi, tide state, pycnocline limits are only collected at tidal stations.
9. The flow value is recorded by Laura Fabian after the sampling run if the station is associated with a USGS Gaging Station. The flow value box is 8 boxes long. The basis, flow value, exponent, & G/L boxes are included.
    a. The basis number refers to whether a flow value is measured or estimated. If the flow value was measured then a 1 goes in the basis box. A 2 is used when the flow value was estimated.
    b. The flow value is a five digit number in cfs (cubic feet per second). If the value given less than five digits then zeros are added following the value to fill all boxes. For example, if the flow value is 261 cfs, then you would enter 26100.
    c. The exponent box denotes the placement of the decimal point for the flow value in the preceding boxes. Following the example above of a flow value of 26100 (for 261 cfs), the exponent would be 3. This exponent denotes moving the

decimal three place from the left, i.e. 261.00 giving the flow value as 261 cfs (the original value).

d. The G/L box is for noting if a value is greater or less than the reported value.
e. For most sampling, the flow values are currently taken from the USGS real-time data website, http://waterdata.usgs.gov/md/nwis/current?type=flow . These values are recorded in 15 minute increments. The increment that most closely matches the sampling time is entered on the field sheet. Note that all data on the website are timed in Eastern Standard Time all year long.

10. The equipment & probe numbers correspond to the number or letter associated with the water quality instrument used to record data. Currently, the Hydrolab instruments have an individual letter associated with them. The instrument is recorded as 9 and then its assigned letter, i.e. 9L.
11. The scientist and senior scientist sign-offs are places for the sampling team to initial, denoting the fact that they collected and verified the data. The spaces are three boxes long. All letters are left justified. If someone only has 2 initials, then they would be placed in the first 2 boxes and the third is left blank.
12. Any comments pertinent to the station or collection of data or samples should be placed in the comments section of the field sheet.

8.2.2 Recording Instrument Readings

1. Remove the storage cup from the sonde.
2. Visually inspect all probes to ensure they are not broken and appear to be in good working order..
3. Install the probe guard.
4. Swirl the instrument in the bucket until the readings stabilize. If using an instrument with a Standard Clark cell, ensure that the water is moving past the DO membrane at 1 ft/sec to obtain an accurate reading.

- Record the water temperature is degrees Celsius. The temperature should be recorded to the nearest tenth.
- Record the pH to the nearest tenth. Place a zero in the hundredths place following the reading.
- Record the dissolved oxygen in mg/L to the nearest tenth. Place a zero in the hundredths place following the reading. The G/L box is available for instances when the DO is reaching its upper or lower limit.
- Record the specific conductance in µS/cm to three significant figures.
- Record salinity in ppt to the nearest tenth.

8.3 Collecting the whole water sample

1. Whole water samples for the Core/ Trend program are collected directly from the bucket. Whole water samples from tidal stations are collected according to the specific SOP for the program being sampled. The sample will either be a one quart or a two quart sample that is sent to DHMH for analysis. One quart is collected for all stations. A second quart is collected for stations sampling for BOD (Potomac core stations). Whole water for the stations including BOD analysis can either be collected in two 1-quart bottles or one 2-quart bottle.
2. Bottles sent to DHMH for analysis must have the station name, date and sample number, e.g. C-12, on the bottle. It is recommended to add "CORE" on the bottle also since some of the same stations are sampled for the Non-Tidal Network Program. Bottles must be labeled legibly with a permanent marker (sharpie). Add depth if multiple depths are being collected at a single station (tidal stations). Do not write on the caps of bottles being sent to the DHMH lab.
3. Stir the water in the sampling bucket (without introducing bubbles) to create a homogeneous sample. Rinse the pre-labeled collection bottle(s) and cap(s) with the sample water 3 times. Fill the bottle(s) and cap tightly.
4. Place the bottle in ice in the sample cooler.
5. The following parameters are analyzed from the whole water bottle: total suspended solids, total alkalinity and turbidity. A second quart is collected to analyze for 5-day BOD on the Potomac Core runs (Monocacy, Mid-Potomac & Lower Potomac runs).
6. Whole water samples for Western Maryland Core follow the above collection procedure, but analysis of certain parameters are completed by the Western Maryland Regional Laboratory. Those parameters are total alkalinity and turbidity. TSS analysis continues to be completed at DHMH in Baltimore.

8.4 Collecting the filtered and particulate samples

8.4.1 Follow the above instructions for filling a bottle for filtration of the sample. Usually a 2- quart bottle or a one quart wide-mouth bottle is used.

8.4.2 Chlorophyll (CHLA)

Chlorophyll samples are not collected for the Western Maryland Core.

1. For each sample, clean a 47mm bell with deionized (DI) water. Set up unit for filtering. Be sure that there is a trap in line between the manifold and the vacuum source.
2. Place a Whatman 47mm GF/F glass fiber filter pad on the filter frit. Always use clean forceps when handling the filter pads.

3. Mix sample thoroughly by agitating and shaking the sample bottle vigorously, then rinse graduated cylinder three times with sample.
4. Agitate the sample again before measuring in the graduated cylinder. Fill graduated cylinder with sample and filter desired volume through filtration unit. Be sure to use a graduate that is close to the volume being filtered (ex: if you are only filtering 80 ml of sample use a 100 ml graduate). Keep the vacuum pressure below 10 inches of Hg (around 8" Hg is good).
5. Filter sufficient volume of sample (20 - 2000 ml) to leave **noticeable color** on the filter pad.
6. Record the total volume filtered on the foil square.
7. Agitate the squirt bottle of $MgCO_3$, as it settles rapidly. Add approximately 1 ml of $MgCO_3$ suspension (1.0 g $MgCO_3$ in 100 ml of DI water) to the last 25 ml of sample in the filtration bell.

NOTE: Samples for dissolved parameters are not to be collected from this filtrate.

8. The pad should be removed as soon as the sample is completely filtered. The pad should not be left on the frit under vacuum. If you are unable to remove it immediately, be sure to release the vacuum to avoid damaging the sample.
9. Using forceps (1 or 2 pair), fold filter in half with sample inside and remove filter pad.
10. Place pad in pre-marked foil square, and carefully fold foil square in thirds, horizontally. Then fold the ends in to seal the filter inside. Be sure forceps do not touch sample residue on the filter pads, because the sample will adhere to the forceps.
11. Be sure that foil square is marked with the date, station, volume of sample filtered, and sample number.
12. Place foil packet into the labeled zip-lock plastic bag and place in the sample cooler on ice.

NOTE: The filter pads for chlorophyll analysis should not be exposed to direct sunlight. Store as soon as possible.

8.4.3 Particulate Carbon/ Particulate Nitrogen (PC/PN)

1. For each sample, clean two 25 mm filter bells with deionized (DI) water.
2. Place a pre-combusted 25 mm GF/F filter (direct from the lab) on each filter frit. Always use clean forceps when handling the filter pads.
3. Mix sample thoroughly by agitating and shaking the sample bottle vigorously, then rinse graduated cylinder three times with sample.
4. Agitate the sample again before measuring in the graduated cylinder. Fill graduated cylinder with sample and filter desired volume through filtration unit.

5. Filter 10-800 ml through each filter. Filter enough sample to leave **noticeable color** on the filter pad.
6. Make sure filter is sucked dry and the **same volume is filtered for both pads**.
7. Record the volume filtered (total volume through one pad - do not add the volumes for the 2 pads together) on the foil square.

NOTE: Samples for dissolved parameters are not to be collected from this filtrate.

8. Using forceps, fold each filter in half.
9. Place both filters in a foil square labeled with date, sample number, station, PC/PN, and volume filtered. Be sure that the pads are not overlapping in the foil square to keep them from freezing together. Fold foil square as described above.
10. Place the folded foil in the labeled zip-lock bag and place in the sample cooler

8.4.4 Particulate Phosphorus (PP)

1. Follow steps above setting up and rinsing two 47 mm filter bells and flasks. The filters used are two Whatman 47 mm GF/F filters (same pads we use for chlorophyll).
2. Filter 50 ml of sample through each filter pad. If 50ml will not go through one pad then filter a smaller volume through multiple pads to attain 50ml of filtrate.
3. Use the filtrate as an equipment rinse and discard.
4. Then filter enough additional (another 20 - 950 ml) to leave **noticeable color** on the filter pad.
5. Record the **total** volume filtered through each pad being sure to add the 50 ml rinse water (total volume through one pad – do not add the volumes for the 2 pads together) on the foil square.
6. **Use this filtrate to fill up the filtrate bottle for the dissolved parameter analysis.**
7. After collecting filtrate, make sure filter is sucked dry.
8. Rinse the filter pad using at least three - 10 ml rinses of DI water, sucking the pad dry after each rinse.

NOTE: If the volume filtered through one pad is ≤ 100 ml, you may want to collect all filtrate in one flask. To do this, filter 50ml to rinse one flask. Collect the remaining volume being filtered in this rinsed flask. Move the filter bell with the unused pad atop the flask containing the filtrate from the first pad (collection flask). Filter and collect the entire volume through the second pad. You may use the second flask as the "pad rinse flask". Place the bell with the already filtered sample pad atop this second flask. Use deionized water to rinse each sample pad 3 times, sucking dry after each rinse.

9. Using forceps, fold each filter in half.
10. Place both filters in a foil square labeled with station, date, PP, sample number, and volume filtered (this is the total volume of

sample through each pad, including the initial 50 ml rinse). Be sure that the pads are not overlapping in the foil square to keep them from freezing together.

11. Fold the foil square as described above. Place foil square in labeled zip-lock bag and place in the sample cooler on ice until you return to the field office.

8.4.5 Filtrate collection (for the dissolved parameters TDN, TDP, NH_4, NO_2+ NO_3, NO_2, PO_4 & DOC)

1. A bottle of filtrate is collected for submission to the DHMH lab for the analysis of dissolved parameters. A 16 oz. bottle is typically used for this purpose. The bottle should be labeled with the station name, date, sample number, e.g. C-12, "CORE" and "filtrate". Bottles must be labeled legibly with a permanent marker (sharpie). Do not write on the caps of bottles going to the DHMH lab.
2. The filtrate should be collected from the PP filtration described in 7.4.4 above.
3. Rinse the labeled bottle and cap 3 times with filtrate.
4. Fill the bottle at least ¾ full with filtrate. Do not fill the bottle above the shoulder.
5. Place the bottle in ice in the sample cooler.

8.5 Completing the Laboratory Sheets

8.5.1 DHMH Chemistry Sheet

1. The chemistry sheets should already be labeled with the station numbers. There should be a separate sheet for each station. The type of sample (whole, filtered, etc) and bottle number should also be pre-filled in. See Appendix V for an example.
2. The following items will need to be filled in:
   - Collector (last names of scientists)
   - Date
   - Start time
   - Field scientist sign-off
   - Start depth (always 0.0 m for bucket samples, but need to be to be filled in at tidal core stations)

8.5.2 DHMH volume Sheet

1. The DHMH volume sheet should have the run name, station names, sample numbers, layer codes and depths all pre-filled in. Depths may need to be added on the volume sheets for the tidal core stations.
2. The following items will need to be filled in
   - Date
   - Time (start time in military time)
   - PP volume filtered, in ml
   - PC/PN volume filtered, in ml
   - Scientist sign-off

8.5.3 CBL Volume Sheet

1. The CBL volume sheet should have the run name, station names, sample numbers, layer codes and depths all pre-filled in. Depths may need to be added on the volume sheets for the tidal core stations.
2. The following items will need to be filled in
   - Date
   - Time (start time in military time)
   - CHLA volume filtered, in ml
   - Salinity, in ppt (for Western MD Core)
   - Scientist sign-off

9.0 Sample Handling and Preservation

1. All samples (whole water, filtrate & pads) must be iced immediately after collection.
2. The whole water and filtrate samples are typically sent to DHMH via a courier. If courier service is used, drain all ice melt water from the cooler and repack, filling with ice up to the necks of the bottles before leaving the cooler at the courier. Be sure that the cooler reads "Baltimore Lab, Water Chemistry, 2th Floor" and "Return to Anne Arundel Health Dept." All lab sheets must accompany the samples in the cooler. Place the completed sheets in the zip-lock bag provided in the field pack.
3. The particulate pads (PC/PN/PP) collected should be kept on ice and placed in the freezer in the bin marked "DHMH" upon return to the Field Office. The volume sheet for the particulate samples should be folded and placed in one of the bags of pads. If you are delivering directly to the DHMH lab then the pads may be given to the lab tech along with the whole water and filtrate bottles and they will store them properly. Samples that have been stored frozen at the Field Office are delivered to DHMH at the end of the sampling week. Use enough ice in the delivery cooler to ensure that the samples stay frozen.
4. Chlorophyll pads are placed in the freezer in the bin marked "CBL" and delivered frozen, directly to the lab on Friday morning.
5. The Western Maryland Core samples are delivered to the Western Maryland Regional Laboratory in Cumberland. The whole water and filtrate bottles are delivered to DHMH in Baltimore via courier. The frozen filters are sent on dry ice directly to the laboratory at a later date, typically on the Thursday after sampling occurs.

10.0 Data and Records Management

10.1 All samples collected are labeled and recorded on the field sheet. The field sheets are reviewed for accuracy and completeness and then submitted to the Principal Investigator and Quality Assurance Officer

(RAS/TEA). A progress report/ cross reference sheet accompanies the original field sheets. The progress report/ cross reference sheet tracks which samples were taken at each site. Any comments or additional samples are noted on this sheet. Notes about instrumentation problems, etc are included with the report. For more information refer to SOP # DR-05: *QA/QC and submission of field data*. An example of a cross reference sheet can be found in Appendix III. Copies of all field sheets, with their attached progress report/ cross reference sheets, are kept on file at the Field Office.

10.2 The whole water and filtered samples submitted to DHMH also have laboratory sheets associated with them. The sheets are submitted with the samples at the time of delivery, by either direct delivery to DHMH or in the cooler via courier. The laboratory data are reported on the lab sheet and sent directly to MDNR, TEA. No copies of the DHMH lab sheets remain at the Field Office.

10.3 The filter pads have a volume sheet associated with them. There is one volume sheet for the particulate pads (PC/PN & PP) that are sent to DHMH. The results for these parameters are submitted to TEA at the bottom of the whole and filtered water sample laboratory sheet. A second volume sheet accompanies the chlorophyll samples to CBL and the results are sent directly to TEA in a data report. No results or lab sheets are kept on file at the Field Office.

11.0 Quality Control and Quality Assurance

11.1 Samples are collected by properly trained staff to ensure continuity of high quality samples. Field staff must adhere to all Standard Operating Procedures.

11.2 Field duplicates are collected every 20 samples to check for accuracy of field collection and preparation of the samples. Laboratory replicates are run every 10 samples.

11.3 Quality control and quality assurance are maintained by proper cleaning and decontamination of sampling gear. Refer to SOP # MC-01: *Cleaning and decontamination of sampling equipment*.

11.4 Equipment blanks of deionized water are submitted on a routine basis by the Non-Tidal Network Program to catch any possible contamination. Refer to SOP MC-02: *Deionized water, blank sample checks*.

11.5 If contamination occurs, every effort is made to pinpoint the source of the contamination and eliminate it.

12.0 References

1. Maryland Department of Natural Resources. 2008. *Quality Assurance Project Plan. Chesapeake Bay Water Quality Monitoring Program- Chemical and Physical Properties Component, 2008-2009.*

2. Maryland Department of Natural Resources. *Standard Operating Procedures Manual.* Water Quality Monitoring Program. WQMP Field Office, 1919 Lincoln Drive, Annapolis, MD 21401.

Appendix A: Core/ Trend Program History

Core History Feb 25. 2009

1974-1997 Bacteriological samples collected all core stations and Potomac Boat.

April 1997 Turkey Pt – CB2.1 (XJH6680) & Sandy Pt – CB3.3C (XHF1373) now being sampled on Main Bay. 15 ft plankton for XHF1373 now sampled @ 5 meters.

May 1998 Bacteriological Labs @ Frederick & Cheverly closed. Monocacy, Mid Potomac, Lower Potomac/ Patuxent and Potomac Boat runs bacti samples dropped.
Bactis still collected for Baltimore, Susquehanna, Hagerstown & Western MD.

April-December 1999 and April, May & September 2000
Only WMD core (no trend) stations sampled due to body shortage at field office.

October 2000
Extra bactis collected @ CCR0001, NBP0103, NBP0534 and NBP0689 for bucket vs. direct stream comparison.

June 2001 name change CHO0626 to ET5.0 (Red Bridges)

July 2002 Western MD samples no longer tested for iron.

November 2003
WMD bacteriological samples no longer submitted. Whole water to WMRL and only filtrate plus pc/pn/pp filter pads to DHMH as of October 2005. Prior to that we were submitting a second quart of water to DHMH that was acid fixed by WMRL and shipped to DHMH via courier.

2004 Remaining Core runs dropped bacteriological sampling.

October & November 2004
Laboratory comparison. Collected regular sampling and 16 ounce filtrate and pc/pn pads at all stations.

July 2005 Began PC/PN/PP & filtered nutrient sampling for all core stations in addition to whole water and chlorophyll pads. Whole water to WMRL and only 16 ounce

filtrate and PC/PN/PP pads to DHMH. Prior to that we were submitting 2 quarts of whole water to DHMH, one of which was acid fixed by WMRL and both were shipped to DHMH via courier

July 2005 PXT0603 (TF1.0). and Kent Narrows (XGG8251) were no longer submitted with the core paperwork. TF1.0 submitted with Patuxent and XGG8251 submitted with Tributary data.

November and December 2008

Extra TSS pads generated with a churn splitter for Monocacy, Susquehanna, Baltimore, Lower Potomac Patuxent and the 2nd day of Western MD core.

January 2009

Chlorophylls now being processed by CBL due to budget cuts and people shortages @ DHMH. Equipment used @ DHMH will be given to CBL to process samples.

April 2011

2 AA vials of filtrate collected for CBL for chloride & sulfate analysis.

November 2011

Enhanced ion matrix samples collected at 6 stations for CBL/ SERC.

September 2012

Discontinued enhanced ion matrix samples

January 2013

Reduced sampling of chloride and sulfate to a subset of stations

October 2013

Samples no longer sent to Western Maryland Regional Lab (WMRL) due to the analyst leaving. All samples now analyzed by DHMH Lab in Baltimore.

July 2015

Total alkalinity and turbidity samples analyzed by WMRL for Western Maryland Core only; TSS, particulate and dissolved parameters continue to be analyzed by DHMH Lab in Baltimore

Appendix B: Progress Report/ Cross Reference Sheet

Maryland Department of Natural Resources
Chesapeake Bay Water Quality Monitoring

Progress Report / Cross Reference Sheet - CORE

Month/ Year: January/ 2009 **Submitted by: Laura Fabian**

Station	Day	Sequence #	Depth (M)	Sample #	Lab (DHMH)	Chloro. (CBL)	Comments
ET5.0 Red Bridges		0901C01	0.0	C-54			
Patuxent Land Core							
PXT0809 Rocky Gorge	7	0901C02	0.0	C-34			
PXT0972 Unity	7	0901C03	0.0	C-35			
Susquehanna Core							
DER0015 Deer Creek	6	0901C04	0.0	C-24			
GUN0258 Glencoe	6	0901C05	0.0	C-27			
GUN0476 Above Prettyboy	6	0901C06	0.0	C-26			
CB1.0 Below Conowingo Dam	6	0901C07	0.0/1	C-25			
			0.0/2	C-25 dup			

Appendix C: Field Sheet

Maryland Department of Natural Resources

Field Sheet

Project Name: Core-Western MD

Submitter: AFO-Fabian

Sequence Number: C38 (punch in 3-9 all cards)

Data

R1	Sampling Station Number	Start Date Year	Start Date Month	Start Date Day	Start time	End Date Year	End Date Month	End Date Day	End Time	Number of Samples	submitter code	Category Code	Total Depth M	Study Code	
	POT2766									01	80	AA		04	0

R2	Sample Method	+/-	Air Temp °C	Tide State	Weather code Yesterday	Weather code Today	Cloud Cover (%)	Wind Direction	Wind Velocity (Knots) Min.	Wind Velocity (Knots) Max.	Secchi (M)	Basis	Flow Value	Exp	G/L	Senior Scientist
	1															

	DO Method	Equip. Set Unit #	Probe Number	Flow Tide Unit #	Wave Height (M)	Upper (M) Pycnocline Limit	Lower (M) Pycnocline Limit	Scientist Sign Off	gage height	
	H	9	9							0

R3 begin comments here

R4 | 0

Rep No.	Depth M	Water +/-	Water Temp. °C	Field pH	Cor. Flag	D O mg/l G/L	D O mg/l	Conductivity G/L	Conductivity Micromhos/cm	Salinity ppt	Rep Code	Sample Depth M	Layer Code	LAB LOGIN Bottle Number	other information
1	00			0	C		0				1	00	S	C-4	

Weather Codes

10 = none
11 = drizzle
12 = rain
13 = rain, heavy
14 = squally
15 = frozen precipitation

pc/pn volume _____ml

pp/tss volume _____ml

USGS # 01610000
Potomac River @ Paw Paw, WV

Date Entered _____

DNR 2/2008

Appendix D: DHMH Chemistry Sheet

R 5 study code: 0 4 Replicate:

Maryland Department of Natural Resources

Sequence Number

6 (punch in 6-13 all cards) 13

Survey: Core: Western MD

Collector: _____

Laboratory Analysis Sheet (Core/Trend Program)

AFO-410-990-4524/L. Fabian

Sample Station Number: P O T 2 7 6 6

Data Code 1D

Date (Year Month Day) _____ Submitter

Start Depth M: 0 0 End Depth M: _____ Start Time: _____ Code: 8 0

Bottle Numbers: _____

Type of Sample: Filtered (2-8 oz. bottles) C - 4 C - 4 Salinity: _____ ppt 0

R 6

Sample Method: 1 Data Category Code: A A Sample Layer: S Field Scientist Sign Off: _____ Date received by Lab (Year Month Day): _____ Time Received by Lab: _____ Batch Number: _____ 0

R 7

Check test Required	Parameter Description / Units	Parameter Code	Method Code	Analysis Problem Code	G/L	Results Record Decimal in a Box	Percent Recovery	Standard Deviation	Number in Sample	Analyst Sign Off
X	TDN as N (F) mg/l	T D N								0
X	Ammonia as N (F) mg/l	N H 4								
X	$NO_2 + NO_3$ as N (F) mg/l	N O 2 3								
X	Nitrite as N (F) mg/l	N O 2								
X	PO_4 as P (F) mg/l	P O 4								
X	Total Dissolved P (F) mg/l	T D P								
X	Dissolved Organic C (F) mg/l	D O C								
	Sulfate as SO_4 mg/l	S O 4								
	Turbidity NTU (W)	T U R B								
	Total Alkalinity (W) mg/l	T A L K								
	Total Susp. Solids (W) mg/l	T S S								
	Dissolved Solids mg/l	D S O L								
X	Part. Phosphorus as P mg/l	P P								
X	Part. Carbon as C mg/l	P C								
X	Part. Nitrogen as N mg/l	P N								

R 8

Date Reported (Year Month Day): _____ Final Lab sign off: _____ QA/QC sign off: _____ Transcriber sign off: _____

Send Results To:

Bruce Michael
DNR D-2 Tawes Building
Annapolis, MD 21401
bmichael@dnr.state.md.us
410-260-8627

0

Date Entered: _____

DNR 5/2005

Appendix E: Particulate Sample Labels

DNR 3/ /2009 DHMH
CAS0479 S
C-7

PP_____ML

DNR 3/ /2009 DHMH
CAS0479 S
C-7

PC/PN_____ML

DNR 1/ /2009 Western MD
PC/PN Blank Core
Place 2 (if you have them left over) pads in foil & submit with rest of samples.

DNR 3/ /2009 DHMH
PAT0176 S
C-18

PP_____ML

DNR 3/ /2009 CBL
PAT0176 S
C-18

CHLA_____ML

DNR 3/ /2009 DHMH
PAT0176 S
C-18

PC/PN _____ML

Appendix F: DHMH Volume Sheet

Baltimore Core DNR-MANTA
DHMH

DATE_____ **SCIENTIST SIGNOFF_____**

STATION	SAMPLE #	LAYER CODE	DEPTH (M)	TIME (MLTY)	PP Vol. Filtered. (ml) Big pads	PC/PN Vol. Filt (ml) Little pads
PAT0176 Patapsco	**C-18**	**S**	**0.0**			
PAT0285 Patapsco	**C-19**	**S**	**0.0**			
GWN0115 Gwynns Falls	**C-20**	**S**	**0.0**			
NPA0165 N BR Patapsco	**C-21**	**S**	**0.0**			
JON0184 Jones Falls	**C-22**	**S**	**0.0**			
GUN0125 Gunpowder Falls	**C-23**	**S**	**0.0**			

Appendix G: CBL Volume Sheet

Baltimore Core

DNR-MANTA
CBL

DATE_____

SCIENTIST SIGNOFF_____

STATION	SAMPLE #	LAYER CODE	DEPTH (M)	TIME (MLTY)	Chlorophyll volume (ml)
PAT0176 Patapsco	C-18	S	0.0		
PAT0285 Patapsco	C-19	S	0.0		
GWN0115 Gwynns Falls	C-20	S	0.0		
NPA0165 N BR Patapsco	C-21	S	0.0		
JON0184 Jones Falls	C-22	S	0.0		
GUN0125 Gunpowder Falls	C-23	S	0.0		

Appendix III: Maryland Department of Health and Mental Hygiene, Environmental Chemistry Division: Standard Operating Procedures for Water Quality Parameters

Table of Contents – Appendix III

Standard Operating Procedure

DETERMINATION OF ALKALINITY BY TITRIMETRY

Standard Method 2320 B

1.0 SCOPE AND APPLICATION

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

1.2 This method is suitable for all concentrations of alkalinity; however, appropriate aliquots should be used to avoid a titration volume greater than 50 mL. The sample must not be filtered, diluted, concentrated, or altered in any way.

1.3 Alkalinity is the acid-neutralizing or buffering capacity of a water body. The alkalinity of many surface waters is primarily a function of carbonate, bicarbonate, and hydroxide content; it is taken as an indication of the concentration of these constituents.

1.4 Measuring alkalinity is important in determining a stream's ability to neutralize acidic pollution from rainfall or wastewater. Alkalinity in excess of alkaline earth metal concentrations is significant in determining the suitability of water for irrigation.

2.0 SUMMARY OF METHOD

2.1 An unaltered sample is titrated to an electrometrically determined end-point of pH 4.5 using an automated system. The sample must not be filtered, diluted, concentrated, or altered in any way.

2.2 Alkalinity as $CaCO_3$ is determined from the volume required of a 0.02 N sulfuric acid (H_2SO_4) to titrate 50 mL of the sample. For samples with high alkalinities that require more than 50 mL of titrant smaller sample volumes are used.

2.3 For samples of alkalinities less than 20 mg/L, the amount of the acid required to reduce the pH exactly 0.30 pH units below pH 4.5 is measured and an extrapolation technique is used to determine the equivalence point.

3.0 INTERFERENCES

Soaps, oily matter, suspended solids, or precipitates may coat the glass electrode and cause a sluggish response. Clean the electrode occasionally. Do not filter, dilute, concentrate, or alter sample.

4.0 HEALTH AND SAFETY

4.1 Good laboratory practices should be followed during reagent preparation and instrument operation.

4.2 The use of a fume hood, protective eyewear and clothing and proper gloves are recommended when handling acids.

4.3 Each employee is issued a *Laboratory Safety Manual* and a *Quality Assurance plan* and is responsible for adhering to the recommendations contained therein.

4.4 Each chemical should be regarded as a potential health hazard. A reference file of material safety data sheet (MSDS) is available in the lab.

5.0 EQUIPMENT AND SUPPLIES

5.1 Equipment

5.1.1 Mantech PC Titration system, consisting of

5.1.1.1 PC-Titrator with Auto-Sampler

5.1.1.2 System Controller with monitor

5.1.1.3 HP Inkjet printer

5.1.1.4 Electrode – Sure-Flow Combination pH electrode, glass body, with BNC connector, Man-Tech # PCE-86-PH1105

5.1.2 Analytical balance – Mettler Toledo AG204

5.2 Supplies

5.2.1 Glass beakers – 100 mL

5.2.2 Graduated cylinder – class A, 50 mL

5.2.3 Volumetric flasks – class A, 50 mL, 100 mL, 500 mL, and 1000 mL

5.2.4 Pipetters – 100 – 1000 µL, 500 – 5000 µL, and 1 – 10 mL

5.2.5 Carboy – 5 L, with spigot, Nalgene

5.2.6 Transfer pipettes – Samco, cat. # 231

5.2.7 pH Electrode filling solution – 4 M KCl saturated with AgCl, Man-Tech # PCE-R001013

6.0 REAGENTS AND STANDARDS

6.1 Reagents

6.1.1 Deionized water

6.1.2 H_2SO_4, 0.02N – Fisher, cat. # SA 226-4

6.2 Standards

6.2.1 pH 4.0 buffer solution – Fisher, cat. # SB 101-500

6.2.2 pH 7.0 buffer solution – Fisher, cat. # SB 107-500

6.2.3 pH 10.0 buffer solution – Fisher, cat. # SB 115-500

6.2.4 Stock standard, 25,000 mg/L $CaCO_3$ (0.5N) – 10 mL/ 16 voluette ampoules, Hach, product # 14278-10

6.2.5 Intermediate standard, 5000 mg/L $CaCO_3$ – Pipet 5 mL of the stock standard (6.2.4) into a 25 mL volumetric flask. Bring to volume with deionized water. Prepare every two weeks.

6.2.6 Check standard, 50 mg/L $CaCO_3$ – Pipet 5 mL of intermediate standard (6.2.5) into a 500 mL volumetric flask. Bring to volume with deionized water. Prepare every two weeks.

6.2.7 Quality control (QC) Sample, QC-MIN-WP – Add approximately 900 mL of deionized water to a 1000 mL volumetric flask. Transfer exactly 10.0 mL of the concentrate from each ampule into the flask. Fill to mark with deionized water. Mix thoroughly. Transfer to a reagent bottle, label, and store at 4 ^{o}C. Prepare every 6 months. If a different QC is used, follow the sample preparation instructions given in the accompanying paper work.

7.0 SAMPLE COLLECTION, PRESERVATION, AND STORAGE

7.1 Samples are collected in 1 liter polyethylene cubitainers and iced or refrigerated to 4 ^{o}C. The holding time is 14 days

7.2 The sample must not be filtered, diluted, concentrated or altered in any way.

8.0 QUALITY CONTROL

8.1 The acceptable range for the slope of the calibration curve is -61 mV to -57 mV. Calibration has to be repeated if the slope falls outside this range.

8.2 A blank and a blank spike are analyzed at the beginning of the run. Blank concentration must be less than the reporting level of 1 ppm and the acceptable value for the spike recovery is 90 – 110%. Blank, blank spike or sample spike not meeting the criteria is reanalyzed.

8.3 Every tenth sample is duplicated and spiked. The acceptable values for the relative percent difference (RPD) are ± 10 and for the spike recovery (SR) are 90 – 110%. If these do not fall within the accepted ranges, the corresponding analyses are repeated.

8.4 A check standard is run after every ten samples. A QC sample is analyzed at the beginning and the end of each analytical run.

8.5 Data acceptance criteria are listed on the data review checklist. (Appendix A).

8.6 Laboratory participates in yearly ERA WatR Supply (WS) and WatR Pollution (WP) Proficiency Tests.

8.7 An initial demonstration of capability study is performed by each analyst performing the test.

9.0 PROCEDURE

9.1 Sample preparation

9.1.1 Prepare a list of samples to be analyzed on a Sample Run Log (Appendix B).

9.1.2 Pour approximately 60 mL of the pH 4, pH 7 and pH 10 buffers into each of the labeled 100 mL beakers.

9.1.3 Pour 50 mL portions of each well mixed sample, measured using a class "A" graduated cylinder, into labeled 100 mL beakers. Pour a duplicate of every tenth sample.

9.1.4 Spike blank and every tenth sample, or one sample per batch if analyzing less than 10 samples, by adding 1 mL of Intermediate standard solution (6.2.5) to 49 mL of deionized water and samples respectively.

9.2 Daily electrode preparation

9.2.1 Rinse the electrode with deionized water to remove crystal residue that may have formed on the surface during storage.

9.2.2 Check the electrolyte level in the reference cavity, which should be approximately ¼ inch below the fill-hole. If the electrolyte level is too low, add filling solution (5.2.7) with a transfer pipet. Replace the cap, and then rinse clean the electrode again.

9.2.3 Remove fill-hole cover during calibration and measurement to ensure uniform flow of filling solution.

9.3 Weekly electrode maintenance

9.3.1 Disconnect the electrode from the unit. Empty the electrode with a transfer pipet. Rinse with deionized water and then, fill up with filling solution. Connect the electrode.

9.3.2 Soak electrode in pH 4 buffer for a minimum of one hour.

9.3.2.1 Follow the steps in 9.4.1 to 9.4.3

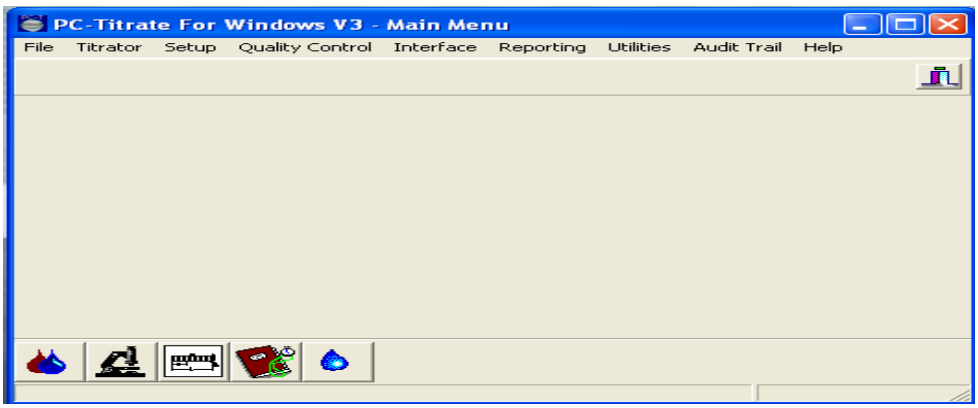
9.3.2.2 Place a beaker with pH 4 buffer in the # 1 position.

9.3.2.3 Select "Tubes" from "Zones", select "1" as the beaker number for "Tubes & the like". Click on "Go to this location XYZ" to send the probe to "1" position.

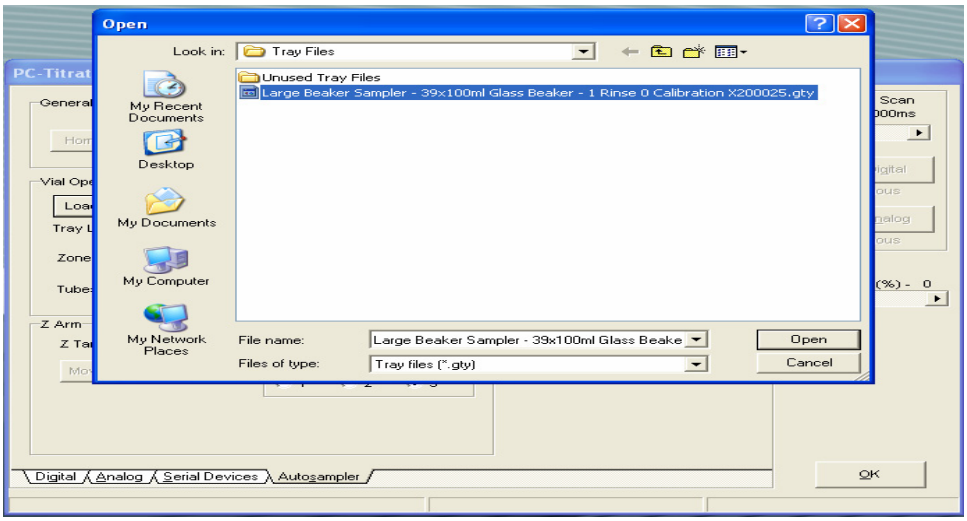
9.4 Instrument preparation

9.4.1 Check and fill the deionized bottle and acid bottle.

9.4.2 Turn on the computer and the autosampler. Double click on "PC-Titrate V3".

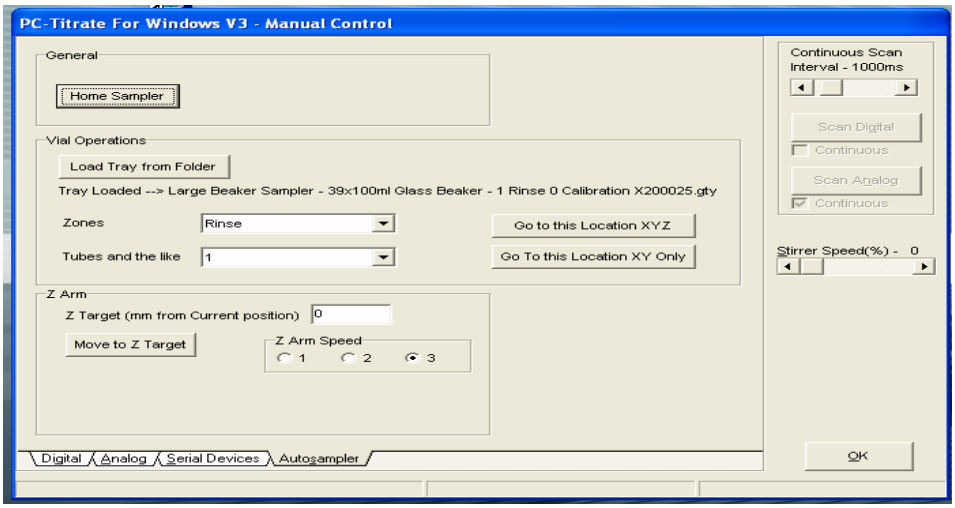


9.4.3 Click on "Titrator" and select "Manual control" from the pull down list. Select "Autosampler", "Load tray from folder", and "Large beaker sampler", and then click on "Open".

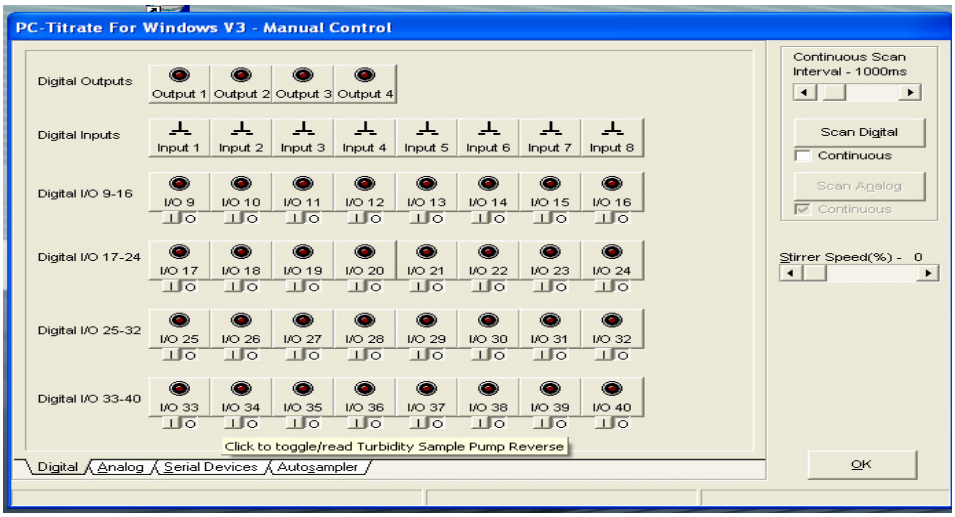


9.4.4 Click on “Home sampler” to send the probe to home position.

9.4.5 Select “Rinse” from “Zones” and “1” as the beaker number for “Tubes & the like”. Click on “Go to this location XYZ” to send the probe to the rinse beaker.



9.4.6 Click on “Digit” tab and “Output 4” to rinse the probe and fill up the beaker. Click “Output 4” again to turn it off.



9.5 Buret preparation

9.5.1 Remove the titrant delivery line from the electrode block on the autosampler and place it into a waste beaker.

9.5.2 Check and fill the acid bottle.

9.5.3 Go to the "Serial devices".

9.5.4 Click on button labeled "Dispense 10%" to dispense the 0.002 N H_2SO_4 through the titrant delivery line. Repeat 2 more times or until no bubbles are observed in the flow.

9.5.5 Fill up the syringe by clicking on "Syringe full down".

9.5.6 Remove the dispenser tip from the waste beaker and return it to its position in the probe holder.

9.6 Electrode calibration

9.6.1 Place pH 4.0, 7.0 & 10.0 buffers into autosampler tray using position # 1, 2 & 3.

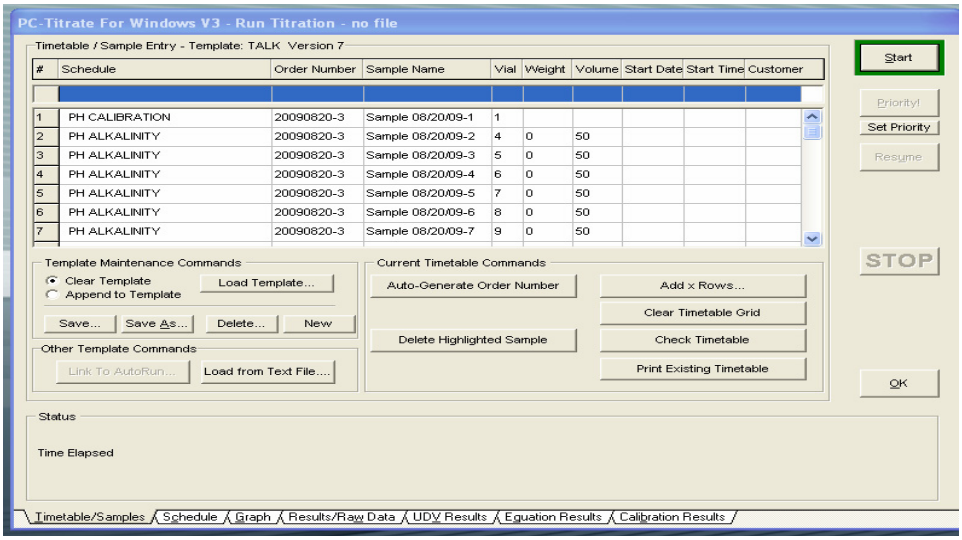
9.6.2 Click on the "Book" tab to open the "pH Calibration" template. Press "Start".

9.7 Sample analysis

9.7.1 Click on the blue "Water Drop" tab to call up the sample table.

9.7.2 Run samples with calibration: Enter "4-7-10" under sample name at the first row reserved for pH calibration and enter 1 in the vial number box.

Enter sample names according to the sample run log (9.1.1) starting with the second row (vial # 4) a check standard, a blank, a blank spike, a QC, and samples to be analyzed. Enter a check standard, a blank, and a QC again at the end of the run.



9.7.3 Highlight each excess line, and then click on "Delete Highlighted Sample" to remove all unused sample information.

9.7.4 Highlight a line and click on "Add x lines" to add additional lines. Left click on the mouse to relocate the lines.

9.7.5 Click "Check Timetable" to verify information entered are valid. Roll down the table to make corrections if needed. Click "OK".

9.7.6 Load the samples according to the run list with the last sample followed by a beaker with the solution recommended by the probe's manufacturer.

9.7.7 Click on "Start".

9.7.8 To run a second tray using the same calibration: Double click on "pH Calibration" and replace it with "pH Alkalinity". Fill in sample names starting with the first row (vial #1). Make sure a set of the quality control samples: check standard, blank, and external QC is also being run at the beginning and at the end in the second tray.

9.7.9 *Calibration Report* and a custom report of *Alkalinity Results* will be printed out automatically at the end of the run.

9.7.10 Recall each titration curve by clicking on "Titrator", "Titration Replay", "Load", and then, selecting date and sample name. Click on "Select" to observe the titration curve. Click "OK" to return to the main menu.

9.7.11 Results can also be printed out by clicking on "Equation results" tab, "Print", and then "OK".

9.7.12 Go to "Manual control" and select "Autosampler" tab. Select "tubes" from "Zones" and "1" as the number for "Tubes & the like". Click on "Go to this location XYZ" to send the probe to the # 1 beaker with the solution recommended by the probe's manufacturer.

9.7.13 Shut down the computer and turn off the autosampler.

10.0 DATA ANALYSIS AND CALCULATIONS

10.1 Alkalinities are calculated automatically by the PC-Titrate V.3 software based on 1 mL of 0.1N H_2SO_4 = 5.0 mg $CaCO_3$

10.1.1 Potentiometric titration to an end point of pH 4.5

$$\text{Alkalinity, mg } CaCO_3/L = \frac{\text{titrant dispensed, mL x 0.02N } (H_2SO_4) \text{ x 50,000}}{\text{sample volume, mL}}$$

10.1.2 Potentiometric titration of low alkalinity

$$\text{Total Alkalinity, mg } CaCO_3/L = \frac{(2B - C) \text{ x 0.02N } (H_2SO_4) \text{ x 50,000}}{\text{sample volume, mL}}$$

where:

B = mL titrant to first recorded pH
C = total mL titrant to reach pH 0.3 unit lower

10.2 Calculate the percentage spike recovery of the laboratory fortified blanks and samples as follows:

$$\%SR = \frac{\text{spiked sample conc. - sample conc., ppm}}{\text{amount of spike added to sample, ppm}} \text{ x } 100$$

10.3 Calculate the relative percentage difference of the duplicated samples as follows:

$$RPD = \frac{\text{difference between the duplicates}}{\text{average of the duplicates}} \text{ x } 100$$

11.0 DATA AND RECORDS MANAGEMENT

11.1 Instrument maintenance, instrument calibration, and quality control data are kept in binders and test results are kept in the file cabinet.

11.2 Normal turnaround time for samples submitted to this lab will be 2 to 10 days from receipt. Results are reported in writing on a sample analysis request form.

11.3 The Division shall retain all pertinent laboratory records for each sample for a period of five years. At the conclusion of this period, clients shall be given the option to take custody of their sample records. The Division shall not be responsible for retrieving, copying, or transferring laboratory records exceeding the five years custody period.

12.0 WASTE MANAGEMENT

12.1 Excess reagents, samples and method process waste are poured into the sink with running water.

12.2 Actual reagent preparation volumes are to reflect anticipated usage and reagent stability.

13.0 REFERENCES

13.1 American Public Health Association, *Standard Methods for the Examination of Water and Wastewater,* Method Number 2320B, 21st Edition, 2005

13.2 Man-Tech Associates inc., *PC-Titrate Windows Software Manual,* version 3.0, November 2004.

13.3 Division of Environmental Chemistry, Maryland Department of Health and Mental Hygiene, *Quality Assurance Plan,* Revision 10.0, 2010

13.4 U.S. Environmental Protection Agency, *Monitoring and Assessing Water Quality, 5.10 Total Alkalinity*, November 2006

13.5 Division of Environmental Chemistry, Maryland Department of Health and Mental Hygiene, *Quality Assurance Plan,* Revision 14.0, 2014

13.6 Division of Environmental Chemistry, DHMH-Laboratories Administration, *Quality Manual,* Revision1.0, November 2014

APPENDIX **A**

Division of Environmental Chemistry
INORGANICS ANALYTICAL LABORATORY

Data Review Checklist – Alkalinity

Standard Methods 2320 B

Lab Numbers[1]:_____

Date Collected: _____ Date Analyzed: _____ Analyst: _____

Procedure	Acceptance Criteria	Status*	Comments
Holding Time	14 days @ 4°C		
Calibration Results	Slope = –61.00 to –57.00 mV		
External QC[2]	Beginning and end of each run		
	Within acceptable range		
Reagent Blank	< Reporting level (1 mg/L)		
Blank Spike	1 per batch		
	Recovery = 90 – 110%		
Check Standard	After every 10th sample and at the end of the run		
	Concentration within 90 to 110% of the true value		
Duplicates/Replicates	Every 10th and the last sample or 1/batch, if less than 10 samples		
	RPD ≤ 10%		
Matrix Spike	Every 10th and the last sample or 1/batch, if less than 10 samples		
	Recovery = 90 – 110%		
Decimal Places Reported	0		
Changes/Notes	Clearly stated		

* Check (√) if criteria are met.

_____ _____
Analyst's Signature & Date Reviewer's Signature & Date

_____
Supervisor's Signature & Date

[2]External QC

Identification = _____
True Value = _____ ppm
Range = _____ ppm

APPENDIX **B**

Division of Environmental Chemistry
INORGANICS ANALYTICAL LABORATORY

Sample Run Log – Alkalinity
Standard Method 2320 B

Date: _____ Analyst: _____

Tray 1 Cup #	Sample ID	Dilution	Tray 1 Cup #	Sample ID	Dilution
1	pH 4		21		
2	pH 7		22		
3	pH 10		23		
4	Ck Std		24		
5	Blank		25		
6	Blank -Spike		26		
7	QC		27		
8			28		
9			29		
10			30		
11			31		
12			32		
13			33		
14			34		
15			35		
16			36		
17			37		
18			38		
19			39		
20					

Tray 2 Cup #	Sample ID	Dilution
1		
2		
3		
4		
5		
6		
7		
8		
9		
10		
11		
12		
13		
14		
15		
16		
17		
18		
19		
20		

Sample Name	Tracking ID
pH 4 Buffer	
pH 7 Buffer	
pH 10 Buffer	
H_2SO_4, 0.02N	

Lab #	Average	RPD	% Spk Rec

Sample Name	Prep Log ID
Intermediate Std, 5,000 ppm	
Ck Std, 50 ppm	
QC:	

[1]Include beginning and ending numbers, account for gaps by bracketing.

[2]QC Sample: _____ Tracking ID: _____

True Value = _____ Acceptable Range = _____

DETERMINATION OF TURBIDITY BY NEPHELOMETRY

EPA Method 180.1

1.0 SCOPE AND APPLICATION

1.1 Turbidity is a principal physical characteristic of water and is an expression of the optical property that causes light to be scattered and absorbed by suspended matter or impurities that interfere with the clarity of the water.

1.2 Determination of turbidity is a common component of water quality assessments. This method is applicable to drinking, ground, waste and saline waters.

1.3 The applicable range of Hach 2100AN Turbidimeter is 0 to 4000 nephelometric turbidity units (NTU). Drinking water samples with turbidity values greater than 40 NTU are diluted and re-analyzed.

2.0 SUMMARY OF METHOD

2.1 This method is based upon a comparison of the intensity of light scattered by the sample under defined conditions with the intensity of light scattered by a standard reference suspension. The higher the intensity of light scattered, the higher the turbidity.

2.2 Readings in NTUs are made using a nephelometer. Detectors of the nephelometer are in place to measure the 90° scattered light, the forward scattered light, the back scattered light and the light transmitted through the sample. The laboratory measures the value in the "Ratio On" mode, in which the instrument's microprocessor uses a mathematical calculation to ratio signals from each detector. The benefits of applying "ratio" on measurements include better linearity, calibration stability, wide measurement range, and the ability to measure turbidity in the presence of color.

3.0 INTERFERENCES

3.1 Etched, scratched, or dirty sample vials or dust contamination within the sample cell compartment and optical compartment scatter light and give inaccurate readings.

3.2 Samples containing air bubbles, coarse debris, or floating sediments can cause erroneous readings.

4.0 HEALTH AND SAFETY

4.1 Good laboratory practices should be followed during inversion of sample and reading of sample result. Use absorbent towels if material is spilled and wash residual into drain.

4.2 Each employee is issued a ***Laboratory Safety Manual*** and is responsible for adhering to the recommendations contained therein.

4.3 Use absorbent towels if material is spilled and wash residual into drain.

4.4 A reference file of MSDS is available in room 7D1.

5.0 EQUIPMENT AND SUPPLIES

5.1 Equipment

5.1.1 Hach Model 2100AN Laboratory Turbidimeter – consisting of a nephelometer with a tungsten-filament lamp for illuminating the sample and detectors to measure scattered light.

5.1.2 Computer – Dell, Microsoft Windows 98

5.1.3 Printer – Hewlett-Parker Deskjet 722C.

5.2 Supplies

5.2.1 Sample cells – 30 mL capacity, item # 20849-00, Hach Co.

5.2.2 Pipettes – Volumetric, class A, 5, 10, 20, and 25 mL.

5.2.3 Flasks – Volumetric, class A, 50 mL, 100 mL and 200 mL

5.2.4 Flasks – Erlenmyer, 50 mL and 100 mL

5.2.5 Gloves – Powder-free, nitrile, item #FF-700, Micro Flex.

5.2.6 Kimwipes – 14.7 x 16.6", item #34721, Kimberly-Clark.

5.2.7 Carboy – 2 ½ gal, with spigot, item # 23210020, Nalgene.

5.2.8 Container – Plastic, for liquid waste, 1 or 2 liter size.

6.0 REAGENTS AND STANDARDS

6.1 Reagents

6.1.1 Deionized water.

6.1.2 Hydrochloric acid, 6N – Fisher Scientific #LC15370-Z.

6.2 Standards

6.2.1 AMCO CLEAR Calibration Kit, for Hach 2100N/AN: 0, 20, 200, 1000, and 4000 NTU – Item # 85525, GFA Chemicals. Use freshly poured portions for calibrating the turbidimeter and discard the used standards

prior to each new calibration. Rinse with DI water and new standard before pouring fresh standards.

6.2.2 AMCO CLEAR Sealed Standards: 0.5, 1.0, 2.0, 5.0, 20, 50, 100, and 200 NTU – Item # 86180, 86443, 86534, 86492, 86122, 85385, 86124, and 86123 respectively, GFA Chemicals. Read these standards at the beginning of each analytical run.

6.2.3 Quality Control Sample – QC-TUR-WS, Spex Certiprep Inc. Empty the entire contents into a small beaker and gently swirl to mix thoroughly. Do not rinse the ampule. Immediately transfer 10.0 mL of the concentrated solution into a 200 mL volumetric flask and bring to volume with deionized water. Mix well and use within 24 hours.

7.0 SAMPLE COLLECTION, PRESERVATION, AND STORAGE

7.1 Samples are collected in liter polyethylene cubitainers and refrigerated or iced to 4 ^{o}C until analysis to minimize microbiological decomposition of solids.

7.2 The holding time is 48 hours when preserved at 4 ^{o}C.

8.0 QUALITY CONTROL

8.1 Instrument Calibration

8.1.1 Primary standards (6.2.1) with concentrations ranging from 0 to 4000 NTU are used to calibrate the turbidimeter every two months.

8.1.2 Sealed secondary standards (6.2.2) with concentrations ranging from 0.5 to 200 NTU are analyzed before each day's run of samples. The instrument check is considered valid when each measured NTU value is within 90 – 110% of its true value. If the values do not fall within the acceptable range the instrument has to be recalibrated using the primary standards (6.2.1) or new standards should be ordered.

8.1.3 AMCO Clear standards are guaranteed to maintain the certified value for 1 year from ship date.

8.2 A mid-range check standard is analyzed after every ten samples and at the end of each run. If a check standard fails, the samples between the last satisfactory check standard and the unsatisfactory standard are re-analyzed.

8.3 Every tenth sample is analyzed in duplicate. The accepted value for the relative percent difference (RPD) is ± 10 %. If the reading does not fall within the accepted ranges, the corresponding analysis is repeated.

8.4 Deionized water is run at the beginning, after every ten samples, and at the end of the run. The accepted value for the blank is less than 0.07 NTU. Routine maintenance includes periodically clean sample cells. Also see Section 9.5.3.

8.5 A quality control sample is analyzed quarterly. Results are kept in a binder next to the instrument.

8.6 A method detection limit (MDL) study is performed once a year by analyzing seven or more replicates of the 0.5 NTU standard spread out through three or more consecutive analytical runs. An MDL study is also performed by each new analyst and when any changes in the analytical procedure are made.

8.7 Data acceptance criteria are listed on the data review checklist (Appendix A).

9.0 PROCEDURE

9.1 Sample Cell Preparation

9.1.1 Clean the samples cells meticulously, both inside and out, and the caps.

9.1.2 Wash the sample cells with soap and rinse with deionized water.

9.1.3 After rinsing, immediately soak the sample cells in a 6N hydrochloric acid solution for a minimum of one hour.

9.1.4 After soaking, immediately rinse the sample cells with deionized water. Rinse a minimum of 15 times.

9.1.5 Immediately after rinsing the sample cells, cap the cells to prevent contamination from the air, and to prevent the inner cell walls from drying out.

9.1.6 Sample cells that are nicked or scratched must be replaced.

9.2 Index New Sample Cells

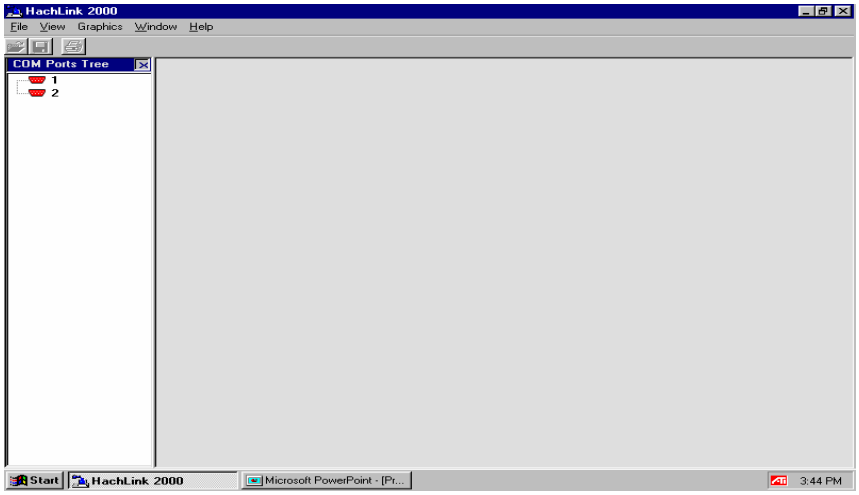
9.2.1 Fill clean sample cells with deionized water to the fill ring mark. Let samples stand for 30 seconds to allow bubbles to rise.

9.2.2 Measure the turbidity at several points of rotation, or as many points as needed, starting with placing the sample cell into the holder with the diamond mark at 6 o'clock position. Mark the orientation where the turbidity reading is the lowest. Use this orientation to perform all sample measurements.

9.2.3 Use the same indexed sample cell, if possible, to measure all the samples.

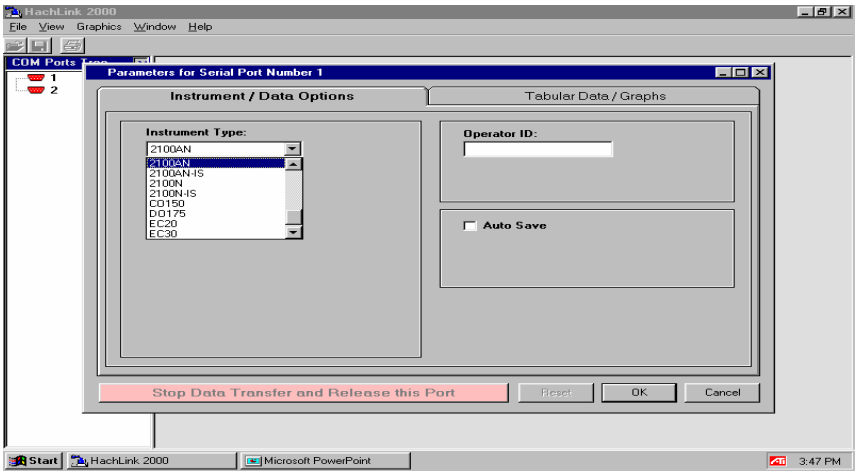
9.3 Instrument Start-up

9.3.1 Leave the turbidimeter on 24 hours a day if the instrument is used daily. Make sure "Ratio", "Sample" and "Signal Average" keys are in "ON" mode displayed by a green light. Maintain "Range" key in "Auto" mode. Select "NTU" from "Units/Exit" key. Turn on the computer. Insert the disk marked as "Turbidity Data". Click on "Hachlink" on the desktop.

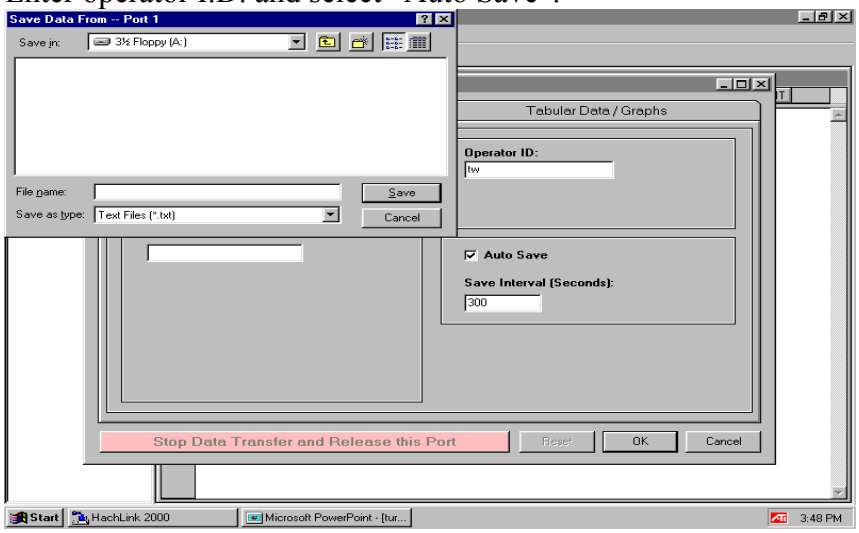


9.3.3 Select "COM Port 1" as the port type by clicking on "1".

9.3.4 Select "2100AN" from the pull down menu of instrument types.

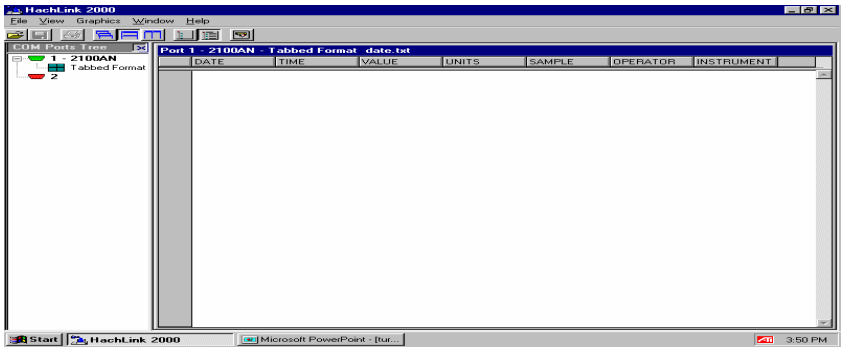


9.3.5 Enter operator I.D. and select “Auto Save”.



9.4 Instrument Calibration

9.4.1 Select “Free Format” for calibration. Enter Date (Cal MM-DD-YY) as file name and click on “Save”.



9.4.2 Press “Cal Zero”. When 00 flashes in green display proceed. Do not shake or mix standards.

9.4.3 Place the “0” NTU standard into the cell holder, align the mark, then close the cell cover.

9.4.4 Press “Enter”. The instrument display counts down from 60 to 0, and then makes a measurement.

9.4.5 The instrument automatically increments to the next standard, 01, as shown on screen in green display. Repeat steps 9.4.3 and 9.4.4 with the rest of the standards: 200, 1000 and 4000 NTU (When the instrument asks for 7500 NTU, press “Cal” to end it.)

9.4.6 Press “Cal Zero” again to store calibration information into memory. Press “Print”. The instrument returns to the sample measurement mode.

9.4.7 Press “Cal” key to review Calibration Data. Use “Δ” key to scroll through the standards. Press the “Print” key prints all of the calibration data in effect. Press the “Units Exit” key to return to the operating mode.

9.4.8 Read sealed secondary standards

9.4.8.1 Follow step 9.3. Select “Tabled Format**”** for sample reading. Enter date as file name. Start with the deionized water as the blank. Thoroughly clean the outside of the sample cell and place it in the sample compartment. Close the sample holder cover.

9.4.8.2 Press “Enter”, then press “Print” to save the reading.

9.4.8.3 Thoroughly clean each of the standard vials. Repeat steps 9.3.8.1 and 9.3.8.2 for all the standards: 0.5, 1.0, 2.0, 5.0, 20.0, 50.0, 100, and 200 NTU.

9.4.8.4 Press “Print”. Keep the printouts in the binder marked “Instrument calibration data”.

9.4.9 Check and fill the carboy with deionized water for rinsing the sample cell when performing sample measurements.

9.5 Sample Analysis

9.5.1 Prepare the list of samples for turbidity on the sample run log sheet (Appendix B) starting with blank, the daily check standards of 0.5, 1.0, 2.0, 5.0, 20, 50, 100, and 200 NTU, the deionized water, then enter each sample number. Measure one replicate, one check standard and one blank for every ten samples. Read check standards again at the end of the run.

9.5.2 Follow step 9.3. Select “Tabled Format**”** for sample reading. Enter date as file name.

9.5.3 Fill the clean and dry glass cell with deionized water. Wipe dry, then insert the cell. If the reading is greater than 0.07 NTU, the cell should be cleaned with detergent and the process repeated. Press “Enter” to clear all previous data, and then press “Print” to transmit data to computer and printer.

9.5.4 Place the 0.5 NTU sealed standard in the sample compartment. Close the cover. Press “Enter” and then press “Print”.

9.5.5 Repeat for the rest of the standards.

9.5.6 Allow samples to reach room temperature to prevent fogging of the cell. Thoroughly mix the sample by gentle inversion. Do not shake. Quickly remove cap and pour approximately 20 ml of sample into the cell for rinse. Immediately fill cell with sample to volume line, wipe dry and insert into turbidimeter. Align the index mark (9.2) on the cell with the raised mark

on the spill ring around the cell holder opening. Be sure the cell has been pushed down completely and is held in place by the spring clip. Close the cover.

9.5.7 Wait for 30 seconds. Check the turbidity reading of the sample from the digital display. Press "Enter", then press "Print" to save the first stable reading at approximately 15 seconds. If the turbidity reading fluctuates, take the cell out, invert to mix well and measure again. Observe the results in the display for accuracy.

9.5.8 Read the rest of the samples according to the run log sheet following step 9.5.6 and 9.5.7. Rinse the cell with deionized water, then rinsed with some of the sample before each sample measurement.

9.5.9 For drinking water sample with turbidities exceeding 40 NTU, dilute the sample with turbidity-free water until turbidity falls below 40 NTU.

9.5.10 After reading all samples, double click the blank area outside the table to go to "Microsoft Excel" table. Enter all sample identifications according to the run log sheet into the sample column. Print out the results.

10.0 DATA ANALYSIS AND CALCULATIONS

10.1 Calculate and report the average for the duplicated samples.

10.2 Multiply sample reading by the dilution factor to obtain the final result for diluted samples.

10.3 Calculate the relative percent difference for the duplicated samples as follows:

$$\text{RPD} = \frac{\text{difference between the duplicates}}{\text{average of the duplicates}} \times 100$$

10.3 All results are reported to one decimal place. The reporting level (RL) is 0.5 NTU. All sample concentrations below this value are recorded as less than 0.5 NTU (< 0.5 NTU).

11.0 DATA AND RECORDS MANAGEMENT

11.1 Normal turnaround time for samples submitted to this lab will be 2 to 10 days from receipt. Results are reported in writing on a sample analysis request form.

11.2 Instrument maintenance, instrument calibration, and quality control data are kept in binders and test results are kept in the file cabinet.

11.3 The Division shall retain all pertinent laboratory records for each sample for a period of five years. At the conclusion of this period, clients shall be given the option to take custody of their sample records. The Division shall not be

responsible for retrieving, copying, or transferring laboratory records exceeding the five years custody period.

12.0 WASTE MANAGEMENT

12.1 It is laboratory's responsibility to comply with all state, federal and local regulations governing waste management, particularly the hazardous waste identification rules and disposal restrictions, and to protect the air, water and land buy minimizing and controlling all releases from fume and bench operation.

12.2 Samples and standards are poured down the drain while large amount of water is running.

13.0 REFERENCES

13.1 United States Environmental Protection Agency, *Methods for Chemical Analysis of Water and Wastes,* Method 180.1 Revision 2.0, August, 1993.

13.2 Hach Company, *Model 2100AN Laboratory Turbidimeter Instruction Manual,* 1993.

13.3 Hach Company Technical Information Series – Booklet No. 11, *Turbidity Science,* 1998.

13.4 The American Public Health Association, *Standard Methods for the Examination of Water and Wastewate,* Method, 21thEdition, 2005.

13.5 Division of Environmental Chemistry, Maryland Department of Health and Mental Hygiene, *Quality Assurance Plan,* Revision 12.0, 2012

13.6 Division of Environmental Chemistry, Maryland Department of Health and Mental Hygiene, *Quality Assurance Manual,* Revision 1.0, 2014

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APPENDIX A
Division of Environmental Chemistry
INORGANICS ANALYTICAL LABORATORY

Data Review Checklist – Turbidity

EPA Method 180.1

Lab Numbers:_____

Date Collected: _____ Date Analyzed: _____ Analyst: _____

Procedure	Acceptance Criteria	Status*	Comments
Holding Time	48 hours @ 4 °C		
Instrument Calibration[2] (0 – 4000 NTU)	Every two months		
Daily Calibration Checks[3] (0 – 200 NTU)	Within 90 to 110% of true values		
Blank	< 0.07 NTU		
Check Standards	After every 10th sample and at the end of the run		
	Concentrations within 90 to 110% of the true values		
Duplicates/Replicates	Every 10th and the last sample or 1/batch of drinking water samples and 1/batch of wastewater samples, if less than 10 samples of each kind		
	RPD ≤ 10 %		
External QC[4] Every two months	Within acceptable range		
	Last date analyzed:		
Decimal places reported	1		
Reporting Level	0.5 NTU; concentrations below this value reported as < 0.5 NTU		
Measured Values	Within range of 0 to 40.0 NTU for drinking water and 0 to 4000 NTU for others		
Diluted Samples	Correct final calculations		
Changes/Notes	Clearly stated		

* Check (√) if criteria are met.

_____ _____
Analyst's Signature & Date Reviewer's Signature & Date

_____
Supervisor's Signature & Date

[1]Include beginning and ending numbers; account for gaps by bracketing.

[2]Sample Name: AMCO CLEAR Calibration kit Tracking ID: _____

[3]Sample Name: AMCO CLEAR Standards Tracking ID: _____

[4]QC Sample: _____ Tracking ID: _____

True Value = _____ Acceptable Range = _____

APPENDIX B

Division of Environmental Chemistry

INORGANICS ANALYTICAL LABORATORY

Sample Run Log – Turbidity

EPA Method 180.1

Date : _____ Analyst: _____

Sample #	Sample ID	Dilution	Conc. NTU
1	Blk (0.0 NTU)		
2	0.5 NTU		
3	1.0 NTU		
4	2.0 NTU		
5	5.0 NTU		
6	20.0 NTU		
7	50.0 NTU		
8	100 NTU		
9	200 NTU		
10	DI Water		
11			
12			
13			
14			
15			
16			
17			
18			
19			
20			
21			
22			
23			
24			
25			
26			
27			
28			
29			
30			

Sample #	Sample ID	Dilution	Conc. NTU
31			
32			
33			
34			
35			
36			
37			
38			
39			
40			
41			
42			
43			
44			
45			
46			
47			
48			
49			
50			
51			
52			
53			
54			
55			
56			
57			
58			
59			
60			

QC Name	Prep Log ID

Lab #	Average	RPD

DETERMINATION OF CHLOROPHYLL BY SPECTROPHOTOMETRY

Standard Method 10200 H

1.0 SCOPE AND APPLICATION

1.1 Phytoplankton (microscopic algae) responds quickly to environmental changes. Some species develop noxious blooms, sometimes creating offensive tastes and odors or anoxic or toxic conditions, and in a very practical sense, they are a part of water quality.

1.2 Chlorophyll *a* concentrations are indicator of phytoplankton abundance and biomass in coastal and estuarine waters.

1.3 This method is applicable to the determination of chlorophyll *a*, *b*, *c* and pheophytin *a* in fresh and marine waters.

2.0 SUMMARY OF METHOD

The chlorophyll and related compounds are extracted from the algae collected on glass fiber filters with aqueous 90% acetone solution. Light absorption of the extract is measured at selected wavelengths and the concentrations of the pigments of interest are calculated using the equations as in 10.0.

3.0 INTERFERENCES

3.1 Chlorophyll solutions degrade rapidly in strong light. Conduct work with chlorophyll extracts in subdued light to avoid degradation. Use opaque containers or wrap with aluminum foil.

3.2 Pheophorbide *a* and pheophytin *a,* two common degradation products of chlorophyll *a*, can interfere with the determination of chlorophyll *a* because they absorb light and fluoresce in the same region of the spectrum as does chlorophyll *a* .

3.2.1 If these pheopigments are present, significant errors in chlorophyll *a* values will result.

3.2.2 Upon acidification of chlorophyll *b*, if present, the resulting fluorescence emission of pheophytin *b* is coincident with that of pheophytin *a*.

3.2.3 Pheophytin *a* is similar in structure to chlorophyll *a*, but lacks the magnesium atom (Mg) in the porphyrin ring. The magnesium can be removed from chlorophyll in the presence of acid.

3.2.4 When a solution of pure chlorophyll *a* is converted to pheophytin *a* by acidification, the absorption peak is reduced to approximately 60% of its original value and shifts from 664 to 665 nm. For pure chlorophyll this before/after acidification absorption peak ratio (OD_{664}/OD_{665}) is 1.7. Solutions of pure pheophytin show no reduction at OD_{665} upon acidification and have 664/665 ratio of 1.0. The *acid ratio* should fall between 1.0 and 1.7. If it is not within this range, the data are not valid and will be discarded.

$$\frac{(OD_{664} - OD_{750})b}{(OD_{665} - OD_{750})a} = \text{acid ratio}$$

b = before acidification
a = after acidification

4.0 HEALTH AND SAFETY

4.1 Good laboratory practice should be followed during reagent preparation and instrument operation.

4.2 The use of a fume hood is recommended when working with acetone.

4.3 Each employee is issued a ***Laboratory Safety Manual*** and is responsible for adhering to the recommendations contained therein.

4.4 A reference file of material safety data sheet (MSDS) is available in lab.

5.0 EQUIPMENT AND SUPPLIES

5.1 Equipment

5.1.1 Spectrophotometer

5.1.1.1 Shimadzu UV-2450PC

5.1.1.2 UVProbe software

5.1.1.3 Computer – Dell T3400 with window 2007

5.1.2 Freezer – Isotemp, flammable-materials storage, cat. # 13-986-425F, Fisher

5.1.3 Tissue Homogenizer – with Con-Torque power unit, cat. # 2355, Eberbach Corp.

5.1.3.1 Chuck for Model 2355 – cat. # 2370, Eberbach,

5.1.3.2 Flexible Coupling for Model 2355 – cat. # 2380, Eberbach

5.1.3.3 Pestle – 30 mL working capacity, cat. # 08-414-14C, Fisher

5.1.4 Centrifuge – Megafuge 2.0, refrigerated, Heraeus Instrument.

5.1.5 Magnetic Stirrer

5.1.6 Refrigerator – 4 ^{o}C, Fisher

5.2 Supplies

5.2.1 Centrifuge tubes – 15 mL graduated conical polypropylene tubes with screw caps, cat. # 05-538-43D, Fisher

5.2.2 Cuvettes – Quartz, 5 cm path, cat. #104-OS 10mm, Hellma USA.

5.2.3 Repipet Dispensers – 5 and 10 mL volume, cat. # 13-687-54 and 13-687-55, Fisher

5.2.4 Transfer Pipets – Disposable Polyethylene, cat. # 13-711-9AM, Fisher

5.2.5 Cuvette Stirrers – cat. # 14-386-22, Fisher

5.2.6 Stirring bar

5.2.7 Beakers – 4000 mL, Pyrex, cat. # 02-555-25K, Fisher

5.2.8 Cylinder – Graduated, 1000 mL, polymethylpentene, cat. # 08-572-5G, Fisher

5.2.9 Reagent bottles – 1 gal size, amber glass jug comes with acetone

5.2.10 Wash bottles – 500 mL, cat. # 03-409-23A, Fisher

5.2.11 Flasks – Class A, volumetric, 100 mL, cat. # 10-210-8C, Fisher

6.0 REAGENTS AND STANDARDS

6.1 Acetone –Spectranalyzed, cat. # A19-4, Fisher

6.2 Aqueous acetone solution, 90% – In a 4 liter beaker, adds 3600 mL of acetone (6.1) and 400 ml of deionized water and mix well.

6.3 Hydrochloric acid, 1N – Certified, cat. # SA48-4, Fisher

6.4 Chlorophyll *a* from spinach or algae – Sigma

7.0 SAMPLE COLLECTION, PRESERVATION, AND STORAGE

7.1 A typical sampling pattern at a station within an estuary includes a vertical series of samples taken from the surface, across the pycnoline, to near bottom, collected at 3 hr. intervals.

7.2 Samples are collected, filtered in the field, sealed in aluminum foils, and labeled. Frozen filtered samples are delivered to the lab as soon as possible and within a week, where samples are ground as soon as possible and within 28 days.

7.3 The acetone extracts are stored in the dark at – 20 ^{o}C and analyzed as soon as possible and within one year.

8.0 QUALITY CONTROL

8.1 90% acetone solution is measured as blank controls at the beginning of the run and for every 10 samples.

8.2 Samples are analyzed in 5 cm cuvettes. Reanalyzed sample with concentration exceeding the calibrated range by reading the sample in 1 cm cuvettes.

8.3 A reference sample, dried Chlorophyll a from spinach or algae (6.4) is analyzed every three months.

8.3.1 Prepare 10 mg/L solution by dissolving the 1 mg of dried spinach chlorophyll *a* in a small amount of 90% acetone in a 100 mL volumetric flask and bring to volume with 90% acetone. Prepare 1 mg/L, 0.1 mg/L, and 0.01 mg/L solutions by serial dilutions.

8.3.2 Analyze the 1 mg/L, 0.1 mg/L, and 0.01 mg/L solutions as regular samples on the spectrophotometer using a 5 cm cuvette with the addition of 3 drop of 1 N HCl.

8.4 Data acceptance criteria are listed on the data review checklist (Appendix A).

9.0 PROCEDURE

9.1 Sample Receiving

9.1.1 Log in and verify the information written on the aluminum pouches of the sample against that listed on the Analysis Request Sheet.

9.1.2 Arrange samples in numerical sequence as they appear on the Analysis Request Sheet and label the stack of pouches with the assigned lab number.

9.1.3 Return labeled samples to the freezer.

9.2 Chlorophyll Extraction

9.2.1 Remove samples from freezer and warm up for 10 minutes at room temperature.

9.2.2 Number centrifuge tubes in numerical sequence. After every ten samples, insert a tube filled with 90% acetone for use as a sample blank.

9.2.3 Place the filter pad in the grinding tube. With a pair of forceps stick the filter 2/3 down to the side of the glass tube.

9.2.4 Turn on the switch of the tissue homogenizer.

9.2.5 Add approximately 4 mL of the 90% acetone to wet the filter. Move tube up and down with the filter tightly against the pestle. Macerate until the whole pad is completely ground up (looks like mush with no pieces of intact pad). A chlorophyll result is only as good as the way it is ground. To avoid spillage, hold tube firmly and move tube slowly.

9.2.6 Add in 10 ml of the 90% acetone to wash down thoroughly the paper pulp and continue to macerate for 30 second.

9.2.7 Pour the homogenate into a labeled 15 mL conical centrifuge tube.

9.2.8 The final volume should be close to 14.5 ml.

9.2.9 Cap the tubes to prevent evaporation of the acetone, cover the whole rack with aluminum foil to avoid light exposure. Store the samples overnight at 4 ^{o}C in refrigerator. Ground samples, prior to centrifugation, are not allowed to stay more than 24 hours.

9.2.10 Take out the samples from refrigerator and centrifuge cold at 3000 rpm for 30 minutes. A pellet smaller than 0.5 mL indicates a good grinding.

9.2.11 Number another set of centrifuge tubes in the same numerical sequence.

9.2.12 Carefully decant the supernatant of each tube to the clean centrifuge tube with the same number.

9.2.13 The clarified extracts are kept in the – 20 ^{o}C freezer until analyzed.

9.3 Logging In Samples for Analysis

9.3.1 Turn on the computer, printer and Shimadzu UV-2450PC.

9.3.2 Open chlorophyll folder in "My Documents". The folder is divided into DNR and MDE chlorophyll files.

9.3.3 Open "Chlorophyll Log in Template".

9.3.4 Click in the left "Header" to enter lab no. and the start date of the sampling. Click in the "Footer" to change the analyst's name.

9.3.5 Enter sample information (sample ID, depth, and volume filtered, etc) starting with one pair of blanks and a pair of blanks after every 10 samples. Label the first blank with the batch's lab number.

9.3.6 Click "Save As" to save the run log into the appropriate folder. Save in "Chlorophyll" folder for appropriate client (MDE or DNR) and the year with a new file as "lab#*chlog*yymmdd"; (My Documents/ Chlorophyll /MDE /Year or My Documents /Chlorophyll /MDE/Year).

9.3.7 Print out the run log and minimize the window.

9.4 Sample Measurement

9.4.1 Click on "UVProbe" on desktop.

9.4.2 Click on "File", and then select "Chlorophyll.pmd" from the "Recent Documents" list. Or look in "Method" folder and select Methods "*.pmd" as file type. Click "Ok".

9.4.3 Maximize the screen and click "Connect" to start the automatic instrument check. Click "OK" when done.

9.4.4 Fill up both cuvettes with 90% acetone and click on "Baseline" to scan from 800 nm to 300 nm.

9.4.5 Name the first blank sample as "Blank-Lab No." under the "Sample ID". Fill the sample cuvette with the extraction blank and click on "Read Unk" (or hit "F9" key). Enter "Blank 1" and repeat the same measurement one more time in the 2nd line.

9.4.6 Enter and read the next 10 samples, before and after acidification for each sample, in the same order as listed in the run log (9.3.6). Indentify and read the 2nd pair of blanks after these 10 samples as "Blank 2" and "Blank 3". All blank samples must have to have a unique name.

Note: Go to “File” and “Save” whenever needed to save the readings in the photometric file.

9.4.7 After the last sample has been analyzed, print out the Sample Table Report.

9.4.8 Go to “File” and click on “Save” to save the data in photometric file. Go to “File” again and click on “Save As”. Select the MDE or DNR chlorophyll folder in the “My Documents”, select “ASCII.*txt “as the file type and type in “Lab#*Chla*yymmdd” as the file name.

9.4.9 Minimize the “UVProbe” window.

9.5 Data Export and Calculation

9.5.1 Open the “ASCII.*txt” file prepared in 9.4.8 above with Excel. .

9.5.2 Highlight the whole area of the 6 wavelength readings for all samples including the blanks under each lab number, right click and select “Copy”.

.

9.5.4 Open the run log prepared in 9.3.6 above.

9.5.5 Click in the most upper left cell that the set of readings is going to be paste to. Right click on the mouse and select “Paste”.

9.5.6 Double check all numbers for correctness after finish.

9.5.7 Click on “Print”.

9.5.8 Click “Save” to save all data in the same “Chlog” file (9.3.6).

10.0 DATA ANALYSIS AND CALCULATIONS

10.1 The chlorophyll *a* and pheophytin *a* concentrations in samples are calculated as follows:

First, 750 nm OD value is subtracted from the reading before and after acidification. Then, corrected values are used in the following equations:

$$\text{Chlorophyll a}, \frac{mg}{m^3} = \frac{26.7\ (OD_{664}b - OD_{665}a)}{V2\ \times L} \times V1$$

$$\text{Pheophytin a}, \frac{mg}{m^3} = \frac{26.7\ [1.7\ (OD_{665}a) - (OD_{664}b)]}{V2 \times L} \times V1$$

Where
V1 = volume of extract in liters
V2 = volume of sample in liters
L = light path length or width of cell in cm
$OD_{664}b$ = optical density corrected for absorbance at 750 nm before acidification
$OD_{665}a$ = optical density corrected for absorbance at 750 nm after acidification

10.2 For reference samples, spinach chlorophyll *a* concentrations are calculated using linear regression as in 8.3.

10.3 Calculate the relative percentage difference for the duplicated samples as follows:

$$RPD = \frac{\text{difference between the duplicates}}{\text{average of the duplicates}} \times 100$$

11.0 DATA AND RECORDS MANAGEMENT

11.2 Copies of the 'Chlog" file with the complete 6 wavelength readings are sent to the customer along with the Analysis Request Sheet.

11.2 Instrument maintenance, instrument calibration, and quality control data are kept in binders and test results are kept in the file cabinet.

11.3 The Division shall retain all pertinent laboratory records for each sample for a period of five years. At the conclusion of this period, clients shall be given the option to take custody of their sample records. The Division shall not be responsible for retrieving, copying, or transferring laboratory records exceeding the five years custody period.

12.0 WASTE MANAGEMENT

The solvent, acetone, is poured into a labeled waste container. Safety officer will arrange periodic pick-up and disposal.

13.0 REFERENCES

13.1 United States Environmental Protection Agency, *Environmental Monitoring and Support Laboratory, Chlorophyll – Spectrophotometric,* March, 1991

13.2 American Public Health Association, *Standard Methods for the Examination of Water and Wastewater,* Method 10200H, 21st Edition, 2005.

13.3 Shimadzu Corporation, UVProbe Tutorial, April, 2008

13.4 Division of Environmental Chemistry, DHMH – Laboratories Administration, *Quality Assurance Plan,* Revision 12.0, October 2012

13.5 Division of Environmental Chemistry, DHMH-Laboratories Administration, *Quality Manual,* Revision 1.0, November 2014

APPENDIX A

Division of Environmental Chemistry
INORGANICS ANALYTICAL LABORATORY

Data Review Checklist – Chlorophyll a

Standard Methods 10200 H

Lab Numbers[1]:_____ Date Collected: _____ Date Received: _____

Date Ground: _____ Date Analyzed: _____ Analyst: _____

Procedure	**Acceptance Criteria**	**Status***	**Comments**
Holding Time	28 days @ –20 °C (filtered pads)		
	1 year @ –20 °C (extract)		
$\frac{OD664b}{OD665a}$	= 1.0 – 1.7		
Reagent Blank (750nm absorbance for 90% Acetone solution)	Beginning and after every 10th sample		
	Absorbance ≤ 0.001		
750nm absorbance before adding acid	≤ 0.007 for DNR samples		
	≤ 0.01 for MDE samples		
Results	Chlorophyll > Pheophytin		
Field Duplicates/Replicates	Calculate RPD		
External QC[2] Analyze Quarterly	Within acceptable range		
	Last date analyzed:		
Changes/Notes	Clearly stated		

* Check (✓) if criteria are met.
[1]Include beginning and ending numbers, account for gaps by bracketing.

_____ _____
Analyst's Signature & Date Reviewer's Signature & Date

_____
Supervisor's Signature & Date

[2]External QC
Identification = _____
True Value = _____ mg/L
Range = _____ mg/L

APPENDIX B
Division of Environmental Chemistry
INORGANICS ANALYTICAL LABORATORY

Sample Run Log – Chlorophyll a

Standard Methods 10200 H

Lab Number: ________________________ Analyst: _____________

Sample ID	**Depth (M)**	**Vol Fil (L)**	**Vol Extr (mL)**	**Cell Ln (cm)**	Before/ After	**750 (nm)**	**665 (nm)**	**664 (nm)**	**663 (nm)**	**647 (nm)**	**645 (nm)**	**630 (nm)**	**Chl-a (µg/L)**	**Pheo (µg/L)**
			14	5	Before									
			14	5	After									
			14	5	Before									
			14	5	After									
			14	5	Before									
			14	5	After									
			14	5	Before									
			14	5	After									
			14	5	Before									
			14	5	After									
			14	5	Before									
			14	5	After									
			14	5	Before									
			14	5	After									
			14	5	Before									
			14	5	After									
			14	5	Before									
			14	5	After									
			14	5	Before									
			14	5	After									

DETERMINATION OF PARTICULATE CARBON & PARTICULATE NITROGEN

Exeter Analytical CE 440

1.0 SCOPE AND APPLICATION

1.1 This method is used to determine the carbon and nitrogen content in organic and inorganic compounds in surface and saline waters.

1.2 This instrument performs elemental analysis of material retained on filters used in water filtration applications.

2.0 SUMMARY OF METHOD

Particulate material on a pre-ignited glass fiber filter is combusted in an oxygen-helium atmosphere at 900°C. The products of combustion are passed over suitable reagents to undergo complete oxidation and removal of undesirable by-products. The remaining gas proceeds to a mixing chamber. This uniform gas mixture, then passes through a series of traps each bracketed with a pair of thermal conductivity detectors. The difference in the signals from each of these pairs of detectors is proportional to the amount of hydrogen (H in H_2O) and carbon (C in CO_2) present in each sample. The only remaining sample gas, nitrogen (N in N_2), is measured against the pure helium carrier gas.

3.0 INTERFERENCES

3.1 Sampling is the single largest determination of data quality. Duplicates or even triplicates sampling is recommended.

3.2 Filter blanks should be treated the same as filter samples in all respects.

4.0 HEALTH AND SAFETY

4.1 Good laboratory practices should be followed during instrument operation.

4.2 Combustion and reduction tubes are heated to 900°C and 700°C respectively. Wear heat resistant gloves and work on a heat resistant bench top when changing these tubes.

4.3 Wear insulated gloves and use tongs to remove hot crucibles from the furnace, and place them on a metal tray.

4.4 Each employee is issued a *Laboratory Safety Manual* and a *Quality Assurance plan* and is responsible for adhering to the recommendations contained therein.

5.0 EQUIPMENT AND SUPPLIES

5.1 Equipment

5.1.1 CE-440 Elemental Analyzer

5.1.2 CEC-490 interface unit

5.1.3 PC computer

5.1.4 Drying oven, 45°C

5.1.5 Muffle furnace

5.1.6 Microbalance, Sartorius ME 5

5.2 Chemicals

5.2.1 Silver Tungstate-Magnesium Oxide on Chromosorb-A, 20 - 30 mesh

5.2.2 Silver Oxide-Silver Tungstate on Chromosorb-A, 20 – 30 mesh

5.2.3 Silver Vanadate on Chromosorb, 20 – 30 mesh

5.2.4 Ascarite, 20 mesh

5.2.5 Magnesium Perchlorate - slightly crush the irregular chunks to approx. 1/16” to 3/32” diameter

5.2.6 Copper wire

5.2.7 Compressed Oxygen gas

5.2.8 Compressed Helium gas

5.3 Supplies

5.3.1 Filters – Whatman GF/F glass fiber, 25 mm diameter, 0.7 μm particle retention

5.3.2 Nickel sleeves – 7 x 5 mm

5.3.3 Tin capsules – smooth, 6 x 2.9 mm

5.3.4 Desiccator

5.3.5 Microspectula – Hayman style, meets ASTM E 124, Fisher cat. no. 21-401-25A

5.3.6 Microforceps – smooth tips

5.3.7 Pinning forceps

5.3.8 Quartz wool

5.3.9 Vacuum grease

5.3.10 Gloves – heat resistant

5.3.11 Crucible dishes – 3" diameter

5.3.12 Crucible tongs

6.0 REAGENTS AND STANDARDS

6.1 Standard

Acetanilide ($C_6H_5NHCOCH_3$), Acros Organics

6.2 External quality control samples

6.2.1 Domestic Sludge – Standard Reference Material 2781, National Institute of Standards & Technology

6.2.2 Marine Sediment Reference Materials (PACS-2) – National Research Council Canada

7.0 SAMPLE COLLECTION, PRESERVATION, AND STORAGE

The filter pads (9.2.1 – 9.2.4) are kept frozen and dried pads (9.2.6) must be desiccated.

8.0 QUALITY CONTROL

8.1 The calibration series must be placed at the beginning of the wheel. (9.3.1)

8.2 Continue the sample run only after the calibration standards have been analyzed and confirmed that the calculated K_C and K_N are acceptable. ($18 \leq K_C \geq 25$, $7 \leq K_N \geq 10$)

8.3 An acetanilide standard and/or a blank should follow each series of ten samples.

8.4 All samples are duplicated. The accepted value for the relative percent difference for field duplicates (RPD) is ± 10 %.

8.5 A standard series (standard, blank) should also be placed at the end of the wheel.

8.6 Data acceptance criteria are listed on the data review checklist (Appendix A).

8.7 A method detection limit (MDL) study is performed once a year by analyzing seven replicates of the PC/PN pads provided by the client. MDL is calculated as follows:

MDL = (t) x (S) where, t = Student's t value for a 99% confidence level
And a standard deviation estimate with n-1 degrees of freedom [t = 3.14 for seven replicates] and S = standard deviation of the replicate analyses.
Each new analyst also performs an MDL study or when any changes in the analytical procedure or instrument are made.

9.0 PROCEDURE

9.1 Preparation for Analysis

9.1.1 Filters – Place the filters in ceramic crucibles/dishes, combust at a temperature of 450 – 500°C for one hour, remove from oven and then place them in a desiccator to be cooled. Remove from the desiccator and store in a closed container. These filters are sent to the field for sample collection.

9.1.2 Nickel sleeves – Place the nickel sleeves in stainless cups and muffle at 900°C for one hour. Remove, cool down in a desiccator, and store in a capped glass jar.

9.2 Sampling, Filtration and Preparation(performed in the field)

9.2.1 Place a pre-combusted filter pad, with rough side up, in a vacuum filtration assembly.

9.2.2 Mix each sample well before pouring a known volume of sample (anywhere from 10 to 500 mL depending on the density of sample) and quickly pour sample into the filtration assembly.

9.2.3 Filter at a low pressure (15 inches Hg); vacuum to dryness and then break the seal of the vacuum.

9.2.4 Fold the filter in half (exposed surface inside), wrap in aluminum foil and label the sample with the date, ID, volume filtered, and scientist signature.

9.2.5 Freeze at –10°C until ready for analysis.

9.2.6 Prior to analysis, samples should be placed in a drying oven at 45°C for at least 12 hours. Once dried, leave samples in a desiccator until ready to analyze.

9.3 Sample Measurements

9.3.1 Prepare a sample run log (Appendix B) starting with a calibration series that is consisting of 1 nickel sleeve blank, 1 condition, 1 tin capsule blank, 1 condition, and then followed with 3 acetanilide standards.

9.3.2 Standard Preparation

9.3.2.1 Weigh out 1200 to 1500 µg of acetanilide into a tin capsule for each standard. For low level samples, choose to use a smaller amount (as low as 500.0 µg) of acetanilide.

9.3.2.2 Quarterly; Weigh out 200 to 250 µg of domestic sludge into a pre-weighed tin capsule as the reference standard for particulate nitrogen (PN).

9.3.2.3 Quarterly; Weigh out about 1000 µg of PACS-2 into a pre-weighed tin capsule as a reference standard for particulate carbon (PC).

9.3.3 Sample Preparation

9.3.3.1 On a clean surface, place a 7 x 5 mm nickel sleeve into the filter loading die with a plastic loading funnel.

9.3.3.2 Fold the filter and squeeze it into the sleeve with a microforceps. Carefully pull out the microforceps. Use the 4 mm loading plunger to force the compressed filter into the nickel sleeve. Make sure no excess filter protrudes above the lip of the sleeve.

9.3.3.3 Transfer the standards and samples into the 64 sample wheel according to the run log (9.2.1).

9.3.4 Instrument Operation

9.3.4.1 On the main menu, click “Run” and select “Carbon, Hydrogen, Nitrogen” in the pull down list. Enter date (ddmmyy) as the run name, then click “Run” to open the sample information box.

9.3.4.2 Enter sample name and sample weight according to the run log. Enter 100 for the weight of the filter samples.

Double check all entries.

9.3.4.3 Click “Run” to open the list of instructions.

9.3.4.4 Installation of the sample wheel

9.3.4.4.1 Open the manual purge valve on the injection box. Loosen the 4 cover screws and lift the lid. Remove the empty wheel if necessary.

9.3.4.4.2 Insert the loaded sample wheel with the locking pin in place. Tilt the wheel slightly, line up the scribe mark on the wheel with the ratchet in the housing, lower the wheel, and make sure that it is properly seated. Place the locking pin in the center hole.

9.3.4.4.3 Close the cover, and tighten equally on all four screws.

9.3.4.4.4 Open and remove any spent capsules in the capsule receiver. Re-install the cover.

9.3.4.4.5 Check the helium pressure to be sure there is adequate gas to perform the run. Adjust the helium pressure to allow for a fill time near 30. The oxygen pressure is set around 25 psi with enough gas available to complete the run. The combustion temperature is set to 900°C, and reduction temperature at 700°C.

9.3.4.4.6 Close the valve. Click “OK” to start the run.

9.3.5 Data Analysis

9.3.5.1 Arrange the data print outs in order.

9.3.5.2 Open the Excel work book template from PCPN / Calculations / Year / Month.

9.3.5.3 Update the sample names and volumes in the spread sheet.

9.3.5.4 Enter the concentrations for PC and PN in their respective columns.

9.5.3.5 Confirm that the calculations are right and the formulae are ok and consistent. Save the file.

9.5.3.9 Double check all entries and print out the results.

9.4 Instrument maintenance

Replace and pack (Appendix C) reduction tube after 400 to 600 runs; combustion tube after 1000 runs; CO_2 or H_2O trap after 500 runs, and helium or oxygen scrubber after 2000 runs.

10.0 DATA ANALYSIS AND CALCULATIONS

10.1 Calculate the concentrations of PC or PN using the following formula:

$$\text{PC/PN, ppm} = \frac{\text{PC/PN, ug}}{\text{sample volume filtered, mL}}$$

10.2 Calculate the relative percent difference (RPD) for the duplicated samples as follows:

11.0 DATA AND RECORDS MANAGEMENT

11.1 Instrument maintenance, instrument calibration, and quality control data are kept in binders and test results are kept in the file cabinet.

11.2 Normal turnaround time for samples submitted to this lab for analysis will be 2 to 10 days from receipt. Results are reported in writing on a sample analysis request form.

11.3 The Division shall retain all pertinent laboratory records for each sample for a period of five years. At the conclusion of this period, clients shall be given the option to take custody of their sample records. The Division shall not be responsible for retrieving, copying, or transferring laboratory records exceeding the five years custody period.

12.0 WASTE MANAGEMENT

All spent capsules, combustion tube, reduction tube, and absorbent tubes are disposed of as regular trash.

13.0 REFERENCES

13.1 Exeter Analytical, Inc., *Model 440 CHN/O/S Elemental Analyzer Manual,* 1994.

13.2 Division of Environmental Chemistry, DHMH – Laboratories Administration, *Quality Assurance Plan,* Revision 14.0, October 2014

13.3 Division of Environmental Chemistry, DHMH-Laboratories Administration, *Quality Manual,* Revision 1.0, November 2014

APPENDIX A

Division of Environmental Chemistry
INORGANICS ANALYTICAL LABORATORY

Data Review Checklist – PC & PN

Exeter Method 440

Lab Numbers[1]:_____

Date Collected: _____ Date Analyzed: _____ Analyst: _____

Procedure	**Acceptance Criteria**	**Status***	**Comments**
Holding Time	28 days @ – 20°C		
2 Acetanilide Calibration Standards	KC = 18 – 25 KN = 7 – 10		
Blank	BC < 500 BN < 250		
Check Standard	After every 10th sample and at the end of the run		
	%C = 71.09 (Range = 70.005 – 71.569) %N = 10.36 (Range = 9.934 – 10.914)		
External PC QC[2] Analyze Quarterly	Within acceptable range		
	Last date analyzed:		
External PN QC[3] Analyze Quarterly	Within acceptable range		
	Last date analyzed:		
Field Filter Blank	PC < 25 µg; PN < 2 µg		
Field Duplicates	RPD Calculated		
Decimal Places Reported	3		
Sample Calculation	Done correctly		
Changes/Notes	Clearly stated		

* Check (√) if criteria are met. [1]Include beginning and ending numbers; account for gaps by bracketing.

_____ _____
Analyst's Signature & Date Reviewer's Signature & Date

_____
Supervisor's Signature & Date

[2]PC QC Sample: _____ Tracking ID: _____

True Value = _____ Acceptable Range = _____

[3]PN QC Sample: _____ Tracking ID: _____

True Value = _____ Acceptable Range = _____

APPENDIX B

Division of Environmental Chemistry
INORGANICS ANALYTICAL LABORATORY

Sample Run Log – Excel Template Particulate Carbon and Particulate Nitrogen

Exeter Method CE 440

Analyst: _____ Date: _____

Position	Sample Name	Sample wt (µg)	C- RESULT	H- RESULT	N- RESULT
1	Blank (B)				
2	cond (%)				
3	Sleeve Blank (B)				
4	cond (%)				
5	STD1 (K)				
6	STD1 (K)				
7	Blank (B)				

Position	Sample Name/No.		Sample wt (µg)	Sample vol (mL)	PC(ug)	PN(g)	ppm PC	ppm PN	avg. PC	avg. PN
8	Acetanilide			-						
9	Filter	a	-	-			ug PC=		ug PN=	
10		b	-	-						
11		a	-							
12		b	-							
13		a	-							
14		b	-							
15		a	-							
16		b	-							
17		a	-							
18		b	-							
19		a	-							
20		b	-							
21		a	-							
22		b	-							
23		a	-							
24		b	-							
25		a	-							
26		b	-							
27		a	-							
28		b	-							
29		a	-							
30		b	-							
31		a	-							
32		b	-							
33	Acetanilide			-			%C=		%N=	
34	Blank		-	-	BC=		BH=		BN=	

APPENDIX C

Division of Environmental Chemistry
INORGANICS ANALYTICAL LABORATORY

Tube Replacement – PC & PN

Exeter Method 440

CHN Mode Combustion Tube

Silver tungstate / Magnesium oxide on chromosorb

Silver Oxide / Silver tungstate on chromosorb

Silver vanadate on chromosorb

Silver gauze

Quartz wool

Platinum gauze

CHN Mode Reduction Tube

Copper wire

Division of Environmental Chemistry
INORGANICS ANALYTICAL LABORATORY

Tube Replacement – PC & PN

Exeter Method 440

CHN Mode Helium & Oxygen Scrubbers

CHN Mode CO_2 Trap

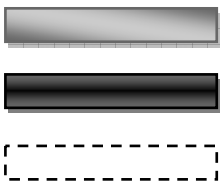
(This end up)

CHN Mode H_2O trap

Ascarite

Quartz wool

Magnesium perchlorate



DETERMINATION OF TOTAL ORGANIC CARBON

Standard Method 5310 B

1.0 SCOPE AND APPLICATION

1.1 This method is applicable to drinking, surface and saline waters, domestic and industrial wastes and provides a speedy and convenient way of determining the degree of organic contamination.

1.2 The fraction of total organic carbon (TOC) that passes through a 0.45 μm pore diameter filter is defined as dissolved organic carbon (DOC).

1.3 The method is applicable to measurement of organic carbon above 0.5 mg/L

2.0 SUMMARY OF METHOD

Organic carbon in a sample is converted to carbon dioxide (CO_2) by catalytic combustion at 680 ^oC. The carbon dioxide formed is purged from the sample, dried, and transferred with a carrier gas to a non-dispersive infrared gas analyzer (NDIR). The amount of carbon dioxide is directly proportional to the concentration of carbonaceous material in the sample.

3.0 INTERFERENCES

3.1 Carbonate and bicarbonate carbons represent interference under the terms of this test and must be removed or accounted for in the final calculation.

3.2 This procedure is applicable only to homogeneous samples which can be injected into the apparatus reproducibly by means of a micro-liter type syringe. The opening of the syringe limits the maximum size of particles which may be included in the following measurements

4.0 HEALTH AND SAFETY

4.1 Good laboratory practices should be followed during reagent preparation and instrument operation. Use of gloves and eye protection is recommended when preparing solutions.

4.2 Each employee is issued a ***Laboratory Safety Manual*** and is responsible for adhering to the recommendations contained therein.

4.3 Each chemical should be regarded as a potential health hazard. A reference file of material safety data sheet (MSDS) is available in lab.

4.4 Gloves, lab coat, and protective eyewear must be used when removing the cover from the furnace and replacing the combustion tube.

5.0 EQUIPMENT AND SUPPLIES

5.1 Equipment

5.1.1 Shimadzu TOC - V_{CPH} Analyzer

5.1.2 Shimadzu ASI-V Autosampler

5.1.3 Computor-Dell Precision T3500

5.1.4 Printer-HP desk jet 990 cxi

5.2 Supplies

5.2.1 Glass vials – 40 mL

5.2.2 Air – Compressed, ultra-zero, UN1002, GTS-Welco

5.2.3 Flasks – Volumetric, 200 mL, 1000 mL

5.2.4 Pipettes – Volumetric, 5 mL, 10 mL, 20 mL, 100 mL

5.2.5 Platinum Catalyst – ST type, P/N 638-60116, Shimadzu Corp.

6.0 REAGENTS AND STANDARDS

6.1 Reagents

6.1.1 Deionized water free from analyte of interest is used to prepare all the reagents and the standards to reduce the carbon concentration of the blank.

6.1.2 2N Hydrochloric Acid – Dilute 166 mL of concentrated Hydrochloric Acid to 1 liter with deionized water.

6.2 Standards

TOC/TIC Standard - custom made standard containing 10 mg/L of total organic carbon and 10 mg/L of total inorganic carbon, cat. # 092 Custom Standard, Environmental Resource Associates.

6.2.1 Potassium Hydrogen Phthalate (KHP) stock standard solution,1000 ppm – Weigh and Stir to dissolve 2.12 g of KHP in about 800 mL of deionized water in a 1 L volumetric flask. Fill to the mark with deionized water. Mix thoroughly. Transfer to a reagent bottle, label, and store at $4^{o}C$. Prepare every month.

6.2.2 KHP working standard, 10 ppm, 20 ppm – Dilute 10 mL and 20 mL of KHP 1000 ppm stock solution to 1 liter in volumetric flasks respectively and mix thoroughly. Transfer to reagent bottles, label, and store at 4 ^{o}C.

6.2.3 KHP working standards – Add 10.0 mL and 100 mL of the 10 ppm stock standard into two 200 mL volumetric flasks respectively. Dilute to mark with water and mix well. This makes working standards of 0.5 mg/L and 5.0, mg/L respectively.

6.2.4 Calibration Standards- By using a vial filled with blank (0 ppm) and a vial of 20 ppm of KHP in the beginning of the tray(see Pg. 6), the instrument automatically prepares the standards of 0.5 ppm, 1ppm, 5ppm, and 10ppm of KHP that will be used for generating the calibration curve.

6.3 Quality Control Sample – An ERA - QC sample with known concentration is analyzed at the beginning and at the end of each tray followed by a blank.

7.0 SAMPLE COLLECTION, PRESERVATION, AND STORAGE

Refrigeration at 4^{o} C prior to analysis is required to minimize microbiological decomposition of solids. The holding time is 48 hours for unacidified samples or 28 days if sample is acidified at the time of collection.

8.0 QUALITY CONTROL

8.1 Reagent grade water is run as the blank control.

8.2 Replicates and spike are performed on every tenth sample or one replicate per run. Duplicated determinations should agree within 10% of their average.

8.3 Spike the sample with 5 ppm KHP by adding 100 μL of 1000 ppm stock solution into 20 mL of the sample. The acceptable spike recovery should be within 10% of the concentration added.

8.4 Quality control (QC) samples including check standard, spiked blank, and external QC (6.3) are analyzed at the beginning and at the end of each run. Each recovery should be within 10% of its true value.

8.5 Instrument check solution, TIC/TOC, is analyzed at the beginning of each run. A reading of 10 ppm of TOC indicates the sample had been properly acidified and inorganic carbon had been successively removed.

8.6 All the standards and samples are analyzed at least three times from each tube. The concentrations reported for the samples are the mean of the triplicates, calculated by the computer program.

8.7 Every tenth sample is duplicated (analyzed from two different tubes) and spiked. The accepted value for the relative percent difference (RPD) or spike recovery is ± 10 %.

8.8 Data acceptance criteria are listed on the Data Review Checklist (Appendix A).

8.9 Laboratory participates in ERA WatR Pollution (WP) Proficiency Test.

9.0 PROCEDURE

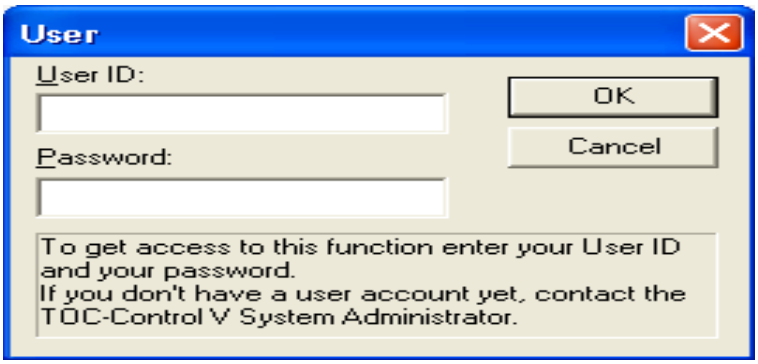
9.1 Analysis Flow

9.1.1 Checking the Dilution Water, Drain Vessel Water, Drain Vessel, and Humidifier water levels- Verify that water volumes are sufficient for analysis. If necessary, replenish water in all bottles.

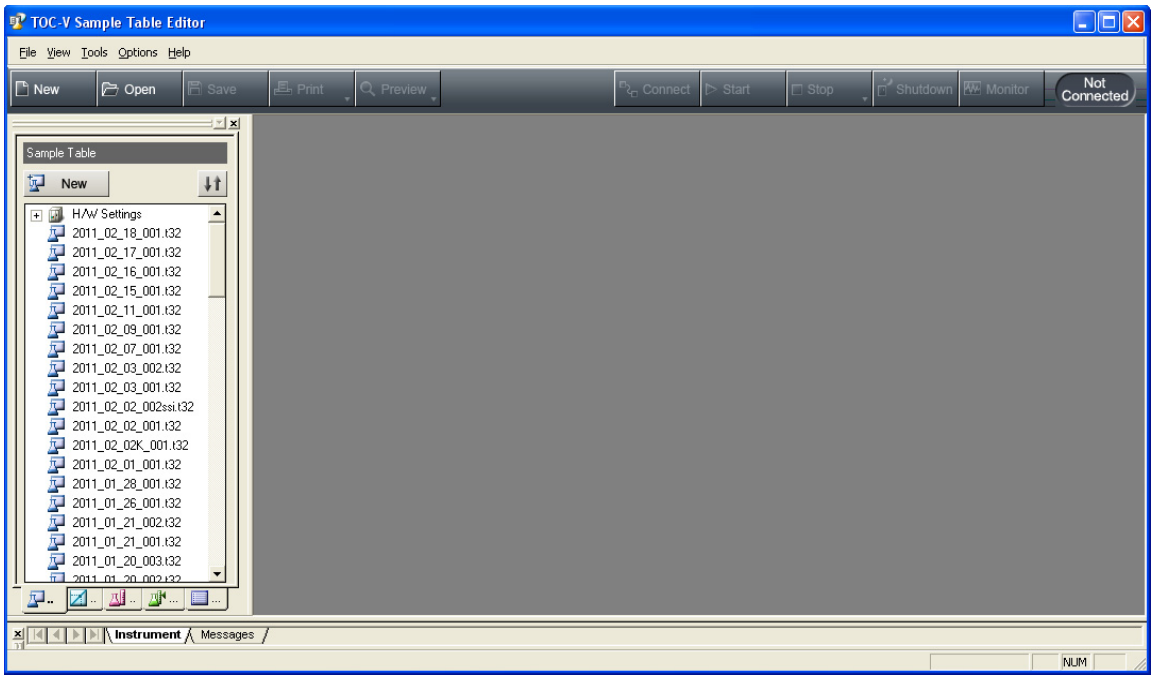
9.1.2 Switch on TOC-V_{cph} Analyzer, which undergoes the initialization sequence.

9.1.3 Turn on the computer.

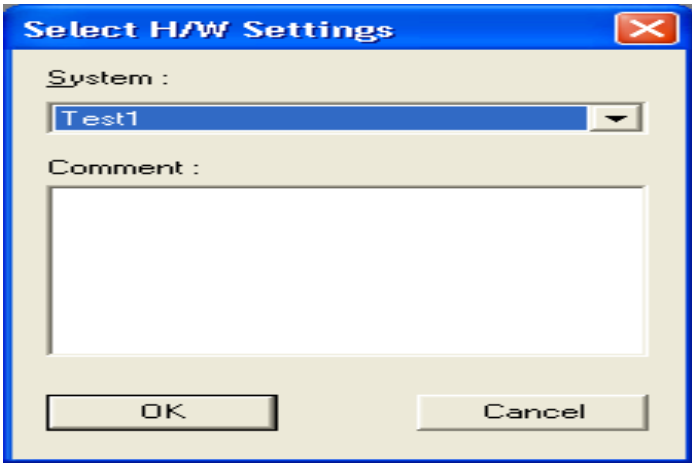
9.1.4 Double click on TOC-V Sample Table Editor icon on the Monitor. The "User" window is displayed.



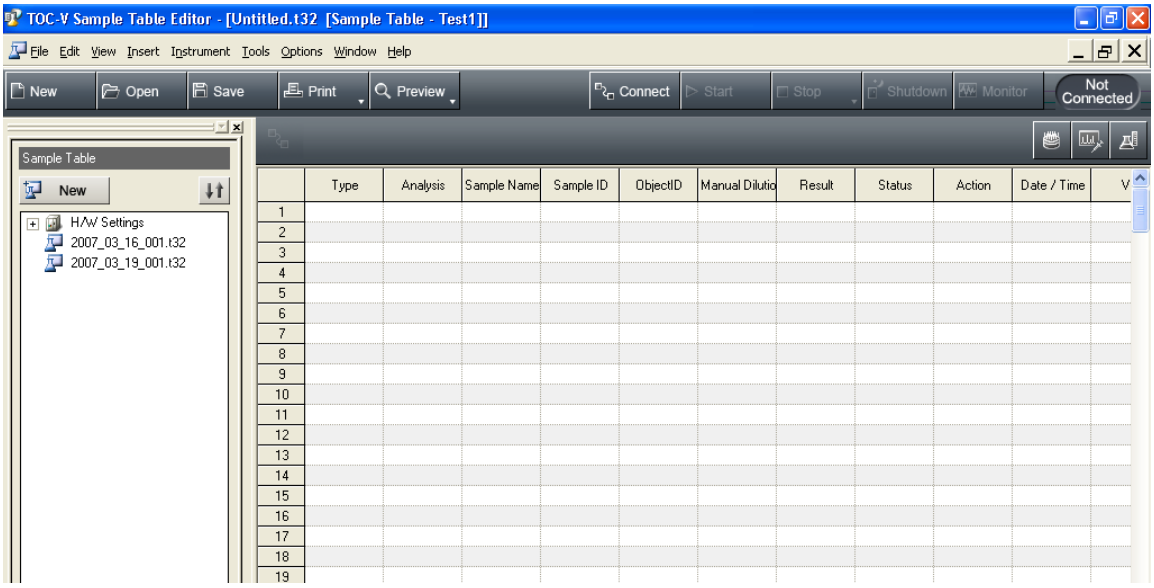
9.1.5 Click OK on the User. A TOC-V Sample Table Editor is opened.



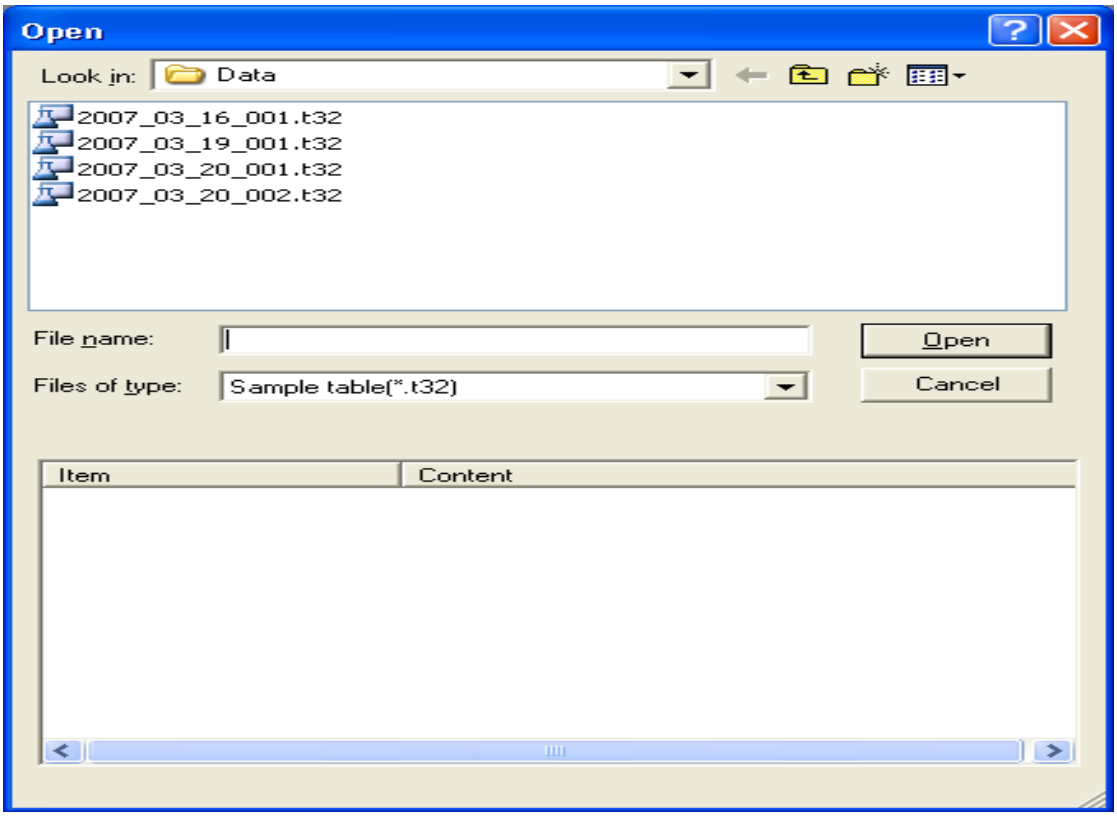
9.1.6 Click “New” on Sample Table Editor, The”Select H/W Settings” window is displayed.



9.1.7 Click OK, an Untitled.t32 [sample table-TOC-V(CPH)] is opened in the Sample Table Editor.

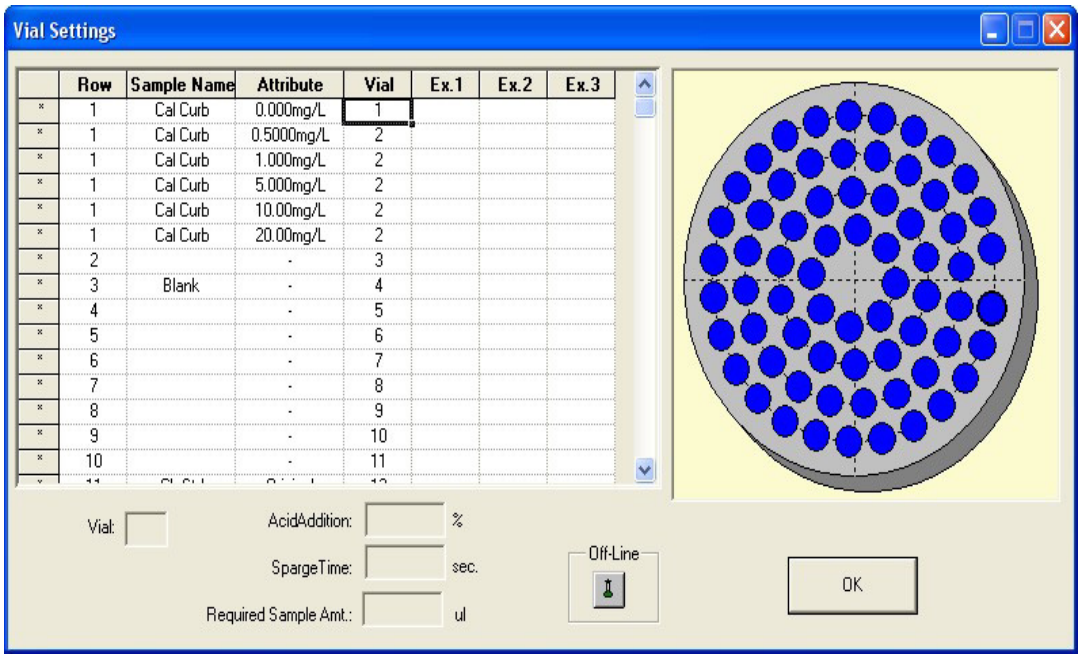


9.1.8 On TOC-V Sample Table Editor, select File, Import Schedule File in the menu Bar. The Open dialog is displayed.



9.1.9 Select the schedule file, and click Open. The schedule content is inserted into the sample table. It demonstrates calibration curve standards, quality control, check standard, blanks, etc.

9.1.10 To enter the vial numbers; enter a number in the sample ID column cell of sample table, click on the lower right part of the cell, and drag downward to enter a series of sample vial numbers all at once. Click on the first row of sample table, click Auto Sample icon, the Vial Setting dialog box is displayed. Select the cell where the last calibration standard is displayed and position the cursor precisely over the lower right corner of the selected cell until "+" is displayed. Click and drag. After all the samples viewed in vial setting, the vial numbers of the vials to be sampled in the ASI-V must be associated with the samples. Click OK in the lower left of "vial Settings."



9.2 Sample Loading

9.2.1 Place the blank vial in positions1 and the Calibration Standard 20ppm in position 2 of the turntable corresponding to the position specified in the ASI condition; i.e. vial 1-2. When automatic injection of acid is specified for NPOC measurement, the bottle containing 2N HCl must be placed next to analyzer.

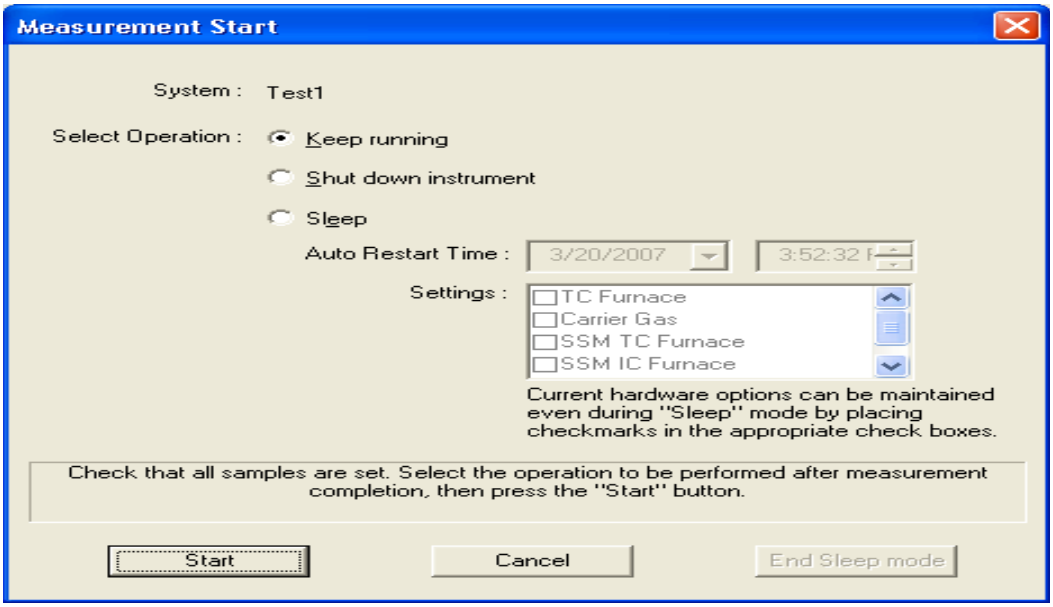
9.2.2 Make a sample run list that starts with the 2 calibration check standards; a blank, the TIC/TOC check, and a QC, followed by sample names with replicates and spikes for every ten samples. A blank, and a check standard are analyzed after every 10 samples. A blank, a check standard and a QC should also be included at the end of each run.

9.2.3 Load sample vials starting with position #1 on the turntable sequence as assigned in the sample worksheet.

9.2.4 The Sample Table must be connected to the connection instrument before analysis can be conducted. Click “Connect” either on the menu bar or click “Instrument-connect.” The icon of the connected instrument and the “Ready” indicator in the Sample Table tab of the file viewer illuminate.

9.2.5 Check that the temperature is reaching 680 ^{o} C and humidifier temperature is 1 ^{o} C. Check the base line at range setting x1 and x50 for position, fluctuation, and noise

9.2.6 Starting the Instrument- Analysis can be started either by clicking “Start” of the menu bar or selecting “Instrument-Start Measurement”. After clicking “Start”, the “Measurement Start” window is displayed. Select the processing that will take place after analysis is complete. Finally, click the start icon at left corner of the Measurement Start window. Analysis starts. When measurement is complete, the results are displayed in the sample table, and analysis ends.



10.0 DATA ANALYSIS AND CALCULATIONS

10.1 Standard curve of 0.0 ppm to 10.0 ppm is established daily and is used directly without shifting to origin assuming the TOC content in water used in preparing standard solution is small enough, with respect to the standard solution concentration, to ignore.

10.2 Calculate % of spike recovery of the laboratory fortified samples as follows

$$\%\,SR = \frac{\text{spiked sample conc. ppm - sample conc., ppm}}{\text{amount of spike added to sample, ppm}} \text{ x } 100$$

10.3 Calculate the % of relative percent difference for the duplicated samples as follows:

$$RPD = \frac{\text{difference of the duplicates}}{\text{average of the duplicates}} \text{ x } 100$$

11.0 DATA AND RECORDS MANAGEMENT

11.1 Instrument maintenance, instrument calibration, and quality control data are kept in binders and test results are kept in the file cabinet.

11.2 Normal turnaround time for samples submitted to this lab will be 2 to 10 days from receipt. Results are reported in writing on a sample analysis request form or through Star Lims for drinking water samples.

11.3 The Division shall retain all pertinent laboratory records for each sample for a period of five years. At the conclusion of this period, clients shall be given the option to take custody of their sample records. The Division shall not be responsible for retrieving, copying, or transferring laboratory records exceeding the five years custody period.

12.0 WASTE MANAGEMENT

12.1 Samples and standards are poured down the drain while flushing with large amount of cold water.

12.2 Actual reagent preparation volumes are to be reflected anticipated usage and reagent stability.

13.0 REFERENCES

13.1 United States Environmental Protection Agency, *Methods for Chemical Analysis of Water and Waste,* Methods 415, August 1993

13.2 American Public Health Association, *Standard Methods for the Examination of Water and Wastewater,* 21st Edition, 2005

13.3 Shimadzu Corporation, *Instrument Manual for Total Organic Carbon Analyzer Model TOC-5000*

13.4 Division of Environmental Chemistry, Maryland Department of Health and Mental Hygiene, *Quality Assurance Plan,* Revision 14.0, 2014

13.5 Division of Environmental Chemistry, DHMH-Laboratories Administration, Quality Manual, Revision 1.0, November 2014……

APPENDIX A

State of Maryland
DHMH - Laboratories Administration
DIVISION OF ENVIRONMENTAL CHEMISTRY
INORGANICS ANALYTICAL LABORATORY

Data Review Checklist – TOC/DOC
Standard Method 5310 B

Lab Numbers[1]:_____

Date Collected: _____ Date Analyzed: _____ Analyst: _____

Procedure	Acceptance Criteria	Status*	Comments
Holding Time	28 days @ – 20 °C for filtered samples; 28 days @ 4 °C for samples acidified to pH < 2 with HCl		
Calibration Curve	Corr. Coeff. ≥ 0.9950		
Sparge Check[2]	TOC = 9 – 11 ppm		
Reagent Blank	< Reporting level (0.50 mg/L)		
Matrix Spike	Every 10th and the last sample or 1/batch, if less than 10 samples		
	Recovery = 90 – 110%		
External QC[3]	Beginning and end of each run		
	Within acceptable range		
Check Standard	After every 10th sample and at the end of the run		
	Concentration within 90 to 110% of the true value		
Duplicates/Replicates	Every 10th and the last sample or 1/batch, if less than 10 samples		
	RPD ≤ 10%		
Decimal Places Reported	2		
Measured Values	Within calibration range (0.50 to10.00 ppm)		
Diluted Samples	Correct final calculations		
Changes/Notes	Clearly stated		

* Check (√) if criteria are met. [1]Include beginning and ending numbers, account for gaps by bracketing.

_____ _____
Analyst's Signature & Date Reviewer's Signature & Date

_____
Supervisor's Signature & Date

[2]Sample Name: TIC & TOC Tracking ID: _____

[3]QC Sample: _____ Tracking ID: _____

True Value = _____ Acceptable Range = _____

APPENDIX B
State of Maryland
DHMH - Laboratories Administration
DIVISION OF ENVIRONMENTAL CHEMISTRY
INORGANICS ANALYTICAL LABORATORY

Sample Run Log –TOC/DOC
Standard Method 5310 B

Date: _____ Analyst: _____

Vial	Sample ID	Dil	Conc. (ppm)	Vial	Lab #	Dil	Conc. (ppm)	Vial	Sample ID	Dil	Conc. (ppm)
1	0-20 ppm		Cal. Std	27				53			
2	0-20 ppm		Cal. Std	28				54			
3	Blank			29				55			
4	Ex QC			30				56			
5	BLK QC			31				57			
6	TOC/TIC			32				58			
7	Blank			33				59			
8	BLK/SPK			34				60			
9	Blank			35				61			
10	MDL			36				62			
11	Blank			37				63			
12	CkStd 5ppm			38				64			
13	Blank			39				65			
14				40				66			
15				41				67			
16				42				68			
17				43				69			
18				44				70			
19				45				71			
20				46				72			
21				47				73			
22				48				74			
23				49				75			
24				50				76			
25				51				77			
26				52				78			

Sample Name	Prep Log ID
KHP Stock Std 1000 ppm	
KHP Std 20 ppm	
KHP Std 10 ppm	
KHP Std 5 ppm	
KHP Std 0.5 ppm	
QC:	

Lab #	Average	%RPD	% Spk Rec

DETERMINATION OF SULFATE BY FLOW INJECTION ANALYSIS

EPA Method 375.2; revision 2.0

1.0 SCOPE AND APPLICATION

1.1 Sulfate (SO_4^{2-}) is widely distributed in nature and may be present in natural waters in concentrations ranging from a few to several thousand milligrams per liter. Mine drainage wastes may contribute large amounts of SO_4^{2-} through pyrite oxidation.

1.2 Sulfur dioxide (SO_2) formed during combustion of fossil fuels containing sulfur can increase the concentration of sulfate in waters during periods of acid precipitation. Sulfur dioxide present in air is converted to sulfuric acid (H_2SO_4) by a number of different ways. The largest sources of sulfur dioxide are electric power generation and industrial facilities.

1.3 The recommended concentration limit for sulfate in drinking water is 250 ppm.

1.4 This automated flow injection analysis method is applicable to drinking and surface waters, domestic and industrial waste waters.

1.5 The applicable range is 10 to 100 mg SO_4^{2-}/L.

2.0 SUMMARY OF METHOD

Sulfate reacts with an ethanolic barium-methyl thymol blue (MTB) solution at a pH of 2.5 to 3.0 and displaces the MTB from the barium to give barium sulfate and uncomplexed MTB. The combined solution is then raised to a pH of 12.5 to 13.0 so that excess barium reacts with MTB. The uncomplexed MTB color is gray and is measured at 460 nm; if it is chelated with barium, the color is blue. Initially, the barium and MTB are equimolar; thus the amount of uncomplexed MTB is equal to the sulfate present.

3.0 INTERFERENCES

3.1 A sodium form cation-exchange column is used to remove the interfering multivalent cations.

3.2 Sample with pH below 2 should be neutralized since high acid concentration can displace cations from the column.

3.2 Sample with pH below 2 should be neutralized since high acid concentration can displace cations from the column.

3.3 Turbid samples should be filtered or centrifuged to eliminate any interference in the colorimetric determination. However, all the samples are filtered in this method to prevent any problems associated with clogging of the small bore tubing and connectors used in Flow Injection Ion Analyzer (FIA).

4.0 HEALTH AND SAFETY

4.1 Good laboratory practices should be followed during reagent preparation and instrument operation. Use of gloves and eye protection is recommended when preparing solutions.

4.2 Each employee is issued a ***Laboratory Safety Manual*** and is responsible for adhering to the recommendations contained therein.

4.3 Each chemical should be regarded as a potential health hazard. A reference file of material safety data sheet (MSDS) is available in the lab.

5.0 EQUIPMENT AND SUPPLIES

5.1 Equipment

Lachat QuickChem 8500 Flow Injection Ion Analyzer, which includes the following:

5.1.1 Automatic sampler ASX-500

5.1.2 Multichannel proportioning pump

5.1.3 Manifold

5.1.4 Colorimetric detector with 460 nm filter

5.1.5 Flow cell- glass with 10 mm path length

5.1.6 Computer with Omnion 3.0 software and Printer

5.2 Supplies

5.2.1 Analytical balance – capable of accurately weighing to the nearest 0.0001 g

5.2.2 Flasks – volumetric, class A, 100, 250, 500, and 1000 mL

5.2.3 Pipettes – volumetric, class A, 5, 10, 20, 25, and 40, 50 mL

5.2.4 Auto-pipetters & tips – ranges from 200 to 1000 uL, 1 to 5 mL, and 2 to 10 mL

5.2.5 Glass column – Hach Company, part # 5000-232

5.2.6 Tubes – 13 x 100 mm disposable culture tubes, Fisher Scientific, cat. # 14-961-27

5.2.7 Glass Vials with caps- 28 x 61 mm, Hach Company, cat. # 21202

5.2.8 Beakers – disposable polyethylene, 50 ml, Fisher Scientific, Cat. # 01-291-10

5.2.9 Whatman Filter papers – folded 114V, 12.5 cm, Fisher Scientific cat. # 1214-125

5.2.10 Ion exchange resin – Zellwegner analytics, cat. # 50233

5.2.11 pH paper with the wide range of 0 to 13.

5.2.12 Graduated cylinders: 25, 50, 100 mL

5.2.13 Plastic Cubitianers (manufactured by Hedwin, available in the warehouse) and 1L Amber glass bottles.

5.2.14 Helium gas: Ultra pure grade

6.0 REAGENTS AND STANDARDS

6.1 Reagents

6.1.1 Deionized water – Used to prepare all the reagents and the standards and as the wash solution.

6.1.2 Carrier, 0.30 mg SO_4^{2-}/L – Dilute 0.3 mL of 1000 mg/L stock sulfate solution (6.2.1) to 1 liter in a volumetric flask.

6.1.3 Barium chloride solution, 6.24 mM – Dissolve 1.526 g of barium chloride dihydrate ($BaCl_2 \cdot 2H_2O$) in about 500 mL of water in a 1L volumetric flask. Dilute to the mark and mix well.

6.1.4 1.0 M Hydrochloric Acid (Carrier/Diluent for Standards) – Add 83.0 mL of concentrated hydrochloric acid (37%, ACS Reagent Grade, d=1.200) to about 800 mL of deionized water in a 1L volumetric flask. Dilute to mark with deionized water after cooling to room temperature. Mix well.

6.1.5 Ethanol (ethyl alcohol) – specially denatured anhydrous alcohol, cat # 245119-2L, Aldrich

6.1.6 Barium-MTB color reagent – Transfer 0.236 g of methylthymol blue (MTB) (3,3-bis-N,N-biscarboxymethyl-amino methylthymol sulfone-

phthalein pentasodium salt) (CASRN 1945-77-3) using 50 mL of barium chloride solution (6.1.3) into a dry 500 mL volumetric flask. Add 4.0 mL of 1M hydrochloric acid solution (6.1.4) and mix. Add 71 mL of water and dilute to the mark with ethanol (6.1.5) and mix well. The pH of this solution should be 2.5. Prepare this solution the day before use and store it refrigerated in a brown plastic bottle.

6.1.7 Sodium hydroxide (NaOH), 50% solution – cat. # SS254-500 Fisher

6.1.8 Working sodium hydroxide, 0.18N Solution – Using a 25 mL graduated cylinder, add 14.4 mL of 50% sodium hydroxide solution (6.1.7) to about 900 mL of water in a 1L volumetric flask. Dilute to the mark and mix well.

6.1.9 Buffered EDTA (for cleaning the Manifold) – Add 6.75 g of ammonium chloride to about 500 ml of DI water in a 1L volumetric flask. Add 57 mL of concentrated ammonium hydroxide and 40g tetrasodium EDTA dehydrate. Mix well and dilute to 1L with distilled water.

6.1.10 Zellwegner analytics ion exchange resin – 50 - 100 mesh for packing the cation exchange column (7.5 Inches long, 2.0 mm ID and 3.6 mm OD Omni fit)

6.1.11 Helium (He) for degassing – Degass the carrier, (6.1.2), barium-MTB color reagent (6.1.6) and the working sodium hydroxide solution (6.1.8) with helium to prevent bubble formation. A pressure of 20 psi for two minutes is recommended.

6.2 Standards

6.2.1 Stock standard, 1000 mg SO_4^{2-}/L – Dry approximately 2 g of sodium sulfate (Na_2SO_4) in aluminum weigh boat at 105 ^{0}C overnight. Cool in a dessicator. Dissolve 1.479 g of dried sodium sulfate in about 800 mL of water in a 1L volumetric flask. Dilute to the mark and mix well.

6.2.2 Calibration Standards – Prepare calibration standards by diluting the stock standard (6.2.1) with water according to the following table. Prepare fresh with each run. DI water is used as 0.0 mg SO_4^{2-}/L (Cal 7) standard.

Standard	Concentration, mg SO_4^{2-}/L	Vol. of 1000 ppm Std (mL)	Final Volume, mL
Cal STD 1	100.0	10.0	100
Cal STD 2	80.0	8.0	100
Cal STD 3	60.0	6.0	100
Cal STD 4	40.0	4.0	100
Cal STD 5	20.0	2.0	100
Cal STD 6	10.0	1.0	100
Cal STD 7	0.00	0.00	100

7.0 SAMPLE COLLECTION, PRESERVATION, AND STORAGE

Samples are collected in one liter polyethylene cubitainers. Samples should be refrigerated or iced at 4°C and should be analyzed as soon as possible to minimize microbiological decomposition of solids. The holding time is 28 days when preserved at 4 °C.

8.0 QUALITY CONTROL

8.1 All the standards and sample are run in duplicates. The concentrations reported for the samples are the averages of the duplicates calculated by the computer program.

8.2 A mid-range check standard, 40 ppm, and a calibration blank is analyzed immediately following daily calibration, at every tenth sample (or more frequently, if required) and at the end of the sample run. If a check standard fails, the samples between the last satisfactory check standard and the unsatisfactory standard are re-analyzed.

8.3 Every tenth sample is duplicated and spiked with 40.0 mg SO_4^{2-}/L (Stock Sodium sulfate solution - 6.2.1). The accepted value for the relative percent difference (RPD) or spike recovery (SR) is ± 10 %. If these do not fall within the accepted ranges, the corresponding analyses are repeated. RPD for samples less than the MDL (lowest calibration standard should not be reanalyzed because they are lower than the detection level and should be considered the same value as the MDL for the calculation.

8.4 A QC sample with a known concentration of sulfate is run at the beginning and at the end of each run. Prepare as recommended by vendor.

8.5 Dilute and reanalyze those samples with a sulfate concentration exceeding the calibrated range.

8.6 Data acceptance criteria are listed on the data review checklist. (Appendix A).

8.7 The laboratory annually participates in ERA water supply (WS) and water pollution (WP) proficiency studies.

8.8 A method detection limit (MDL) study is performed annually by analyzing seven replicates of the 10.00 mg SO_4^{2-}/L standard spread out through three consecutive analytical runs. An MDL study is also performed by each new analyst and when any changes in the analytical procedure are made.

8.9 A demonstration of capability study is performed by each analyst performing the test. Required is the method's MDL study, four replicates of an external quality control sample (2 runs), four blanks and four blank spikes and four matrix spikes analyzed during four analytical runs.

9.0 PROCEDURE

9.1 Sample Preparation

9.1.1 Make a list of the samples to be analyzed for sulfate (Appendix B).

9.1.2 Gravity filter 20 to 25 mL aliquots of samples using Whatman 114V filters into labeled 50 ml disposable polypropylene beakers. Transfer the filtered samples into labeled 13x100 mm tubes.

9.1.3 Prepare spiked sample by adding 400 µL of 1000 ppm stock standard to 9600 µL of the filtered sample.

9.2 Instrument Preparation

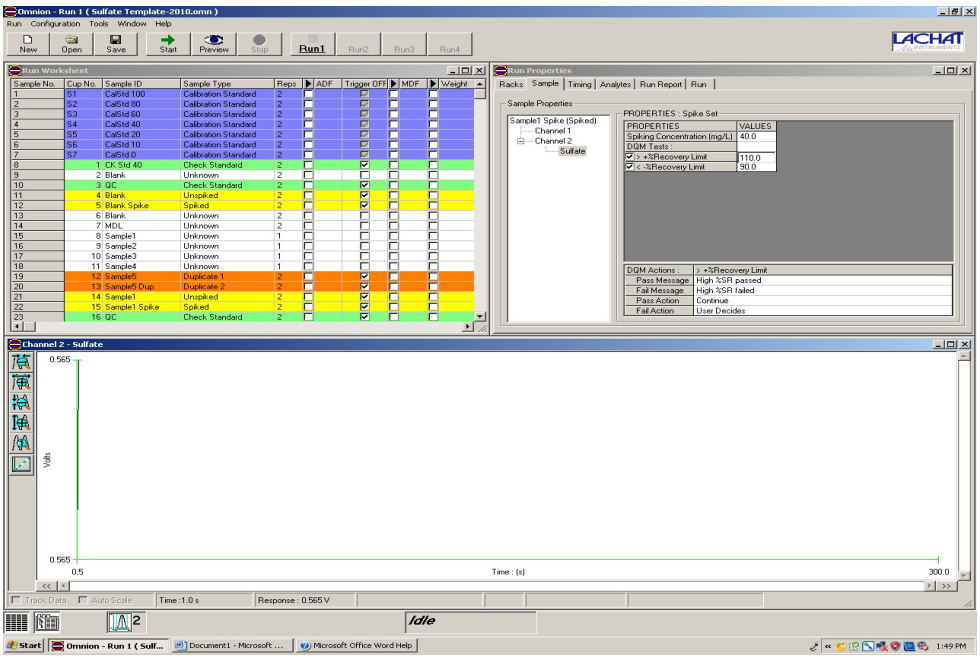
9.2.1 Power up the instrumentation. Inspect all modules for proper connections.

9.2.2 Fill the carrier, well and the reagent rinse bottles with deionized water. Place the lines into the proper water bottle. Raise tension levers on pump tube cassettes.

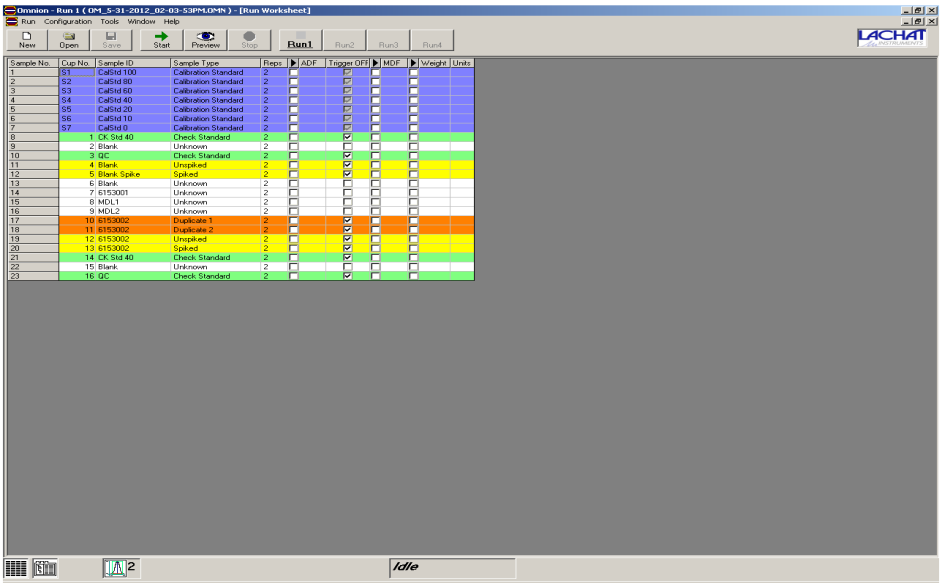
9.2.3 Turn the column off. Turn the pump on. Set the speed at 35 and run deionized water through all the lines for 30 minutes. Check for leaks and smooth flow.

9.3 Run Set-up

9.3.1 Turn the computer on. Double click on "**Omnion 3.0**". Click on "**Open**" and select the Sulfate template. The template is filed in the current year's data folder located in Omnion / Data / methods / sulfate / 2012 (current year).



9.3.2 Set up the Run Worksheet with the standards and samples to be run according to the run log (9.1.1). Go to "**Run**" and click on "**Export Work Data**" to print out the worksheet.

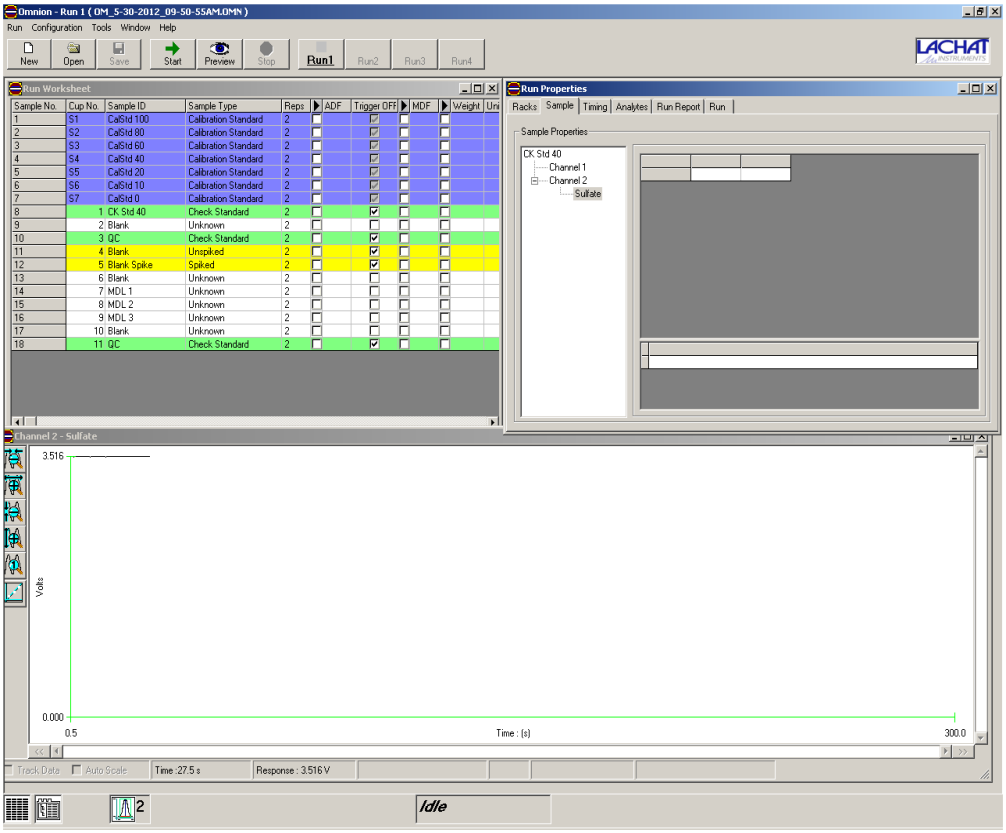


9.4 Sample Analysis

9.4.1 Fill up the water bottle and rinse bottle again with deionized water. Make sure that the carrier and the reagents have been degassed prior to analysis. Pour the carrier, NaOH and the color reagent into the proper bottles. Place the reagent feed lines into the appropriate bottles. With the column still off, start to run the reagents through the system for 10

minutes (or until a stable baseline is attained). Then, turn the column on (9.6) and continue pumping for about 10 minutes before starting the run.

9.4.2 Pour calibration standards and QC into labeled 13x100 mm tubes and place the standards in the cup # 1 to 7 in descending order of concentration. Place sample tubes in the sample holders on the sample rack. Click on preview to monitor the baseline.



9.4.3 Once the baseline is established, click on "**Stop**" to stop monitoring the baseline. Start the analysis by pressing "**Start**" to begin the run template.

9.4.4 If calibration passes, the instrument will continue to analyze the samples. However, if it fails, take appropriate corrective action and recalibrate the instrument.

9.4.5 At the end of the run, click on "**Tools**" and "**Custom**" to print out the custom report.

9.4.6 At the end of each day, the system should be washed with the buffered EDTA solution (6.1.9). This is done by placing all lines in reagent water for a few minutes. **Turn off the column** and then place the Barium-MTB color reagent line and the sodium hydroxide line in the buffered

EDTA solution for 10 minutes. Wash the system with reagent water for 15 minutes and air dry the lines before shutting down.

9.4.7 Turn off the pump, all modules and release levers on pump tube cassettes.

9.5 Troubleshooting

9.5.1 Baseline

9.5.1.1 Make sure that all the reagents are degassed.

9.5.1.2 Check if air is being introduced to the system.

9.5.1.3 Check for any air bubbles in the cation exchange column. Repack the column if necessary.

9.5.1.4 Check for clogged manifold components.

9.5.2 Poor Precision

9.5.2.1 Clean the valve by rinsing all six ports with deionized water. Check and replace if necessary the flared tubing. It may be necessary to return the unit to the factory for valve alignment.

9.5.2.2 Inspect all pump tubes for wear and replace as required.

9.5.2.3 Check for flow restrictions in the manifold.

9.5.2.4 Check for flow restrictions in the column. If large number of dirty samples have been run through the column, it may be necessary to repack the column. It is recommended that dirty samples are analyzed at the end of the run.

9.5.2.5 Replace the sample loop if it is dirty or clogged.

9.6 Column Preparation

9.6.1 Prepare approximately 0.5 g of resin (6.1.10) by mixing it with sufficient water to make a slurry.

9.6.2 Remove one end fitting from the glass column. Fill the column with water then aspirate the slurry or allow it to settle by gravity to pack the column.

9.6.3 Take care to avoid trapping air bubbles and to remove any resin particles from the threads of the glass before replacing the end fitting.

10.0 DATA ANALYSIS AND CALCULATIONS

10.1 Calculate % of spike recovery of the laboratory fortified samples as follows:

$$\%SR = \frac{\text{spiked sample conc. - sample conc., ppm}}{\text{amount of spike added to sample, ppm}} \times 100$$

10.2 Calculate the % of relative standard deviation for the duplicated samples as follows:

$$RPD = \frac{\text{difference of the duplicates}}{\text{average of the duplicates}} \times 100$$

10.3 The MDL is calculated by using seven replicates of the lowest standard solution and is equal to the **standard deviation x 3.143**.

11.0 DATA AND RECORDS MANAGEMENT

11.1 The analysis data of the calibration blank, external QC, MDL and Instrument Performance Check Solution (IPC), i.e. the mid-range check standard, is kept on file with the sample analyses data.

11.2 Normal turnaround time for the analysis of samples submitted to this lab will be 2 to 10 days from receipt. Results are reported in writing on a sample analysis request form.

11.3 The Division shall retain all pertinent laboratory records for each sample for a period of five years. At the conclusion of this period, clients shall be given the option to take custody of their sample records. The Division shall not be responsible for retrieving, copying, or transferring laboratory records exceeding the five years custody period.

12.0 WASTE MANAGEMENT

12.1 It is laboratory's responsibility to comply with all state, federal and local regulations governing waste management, particularly the hazardous waste identification rules and disposal restrictions, and to protect the air, water and land buy minimizing and controlling all releases from fume and bench operation.

12.2 Samples and standards are poured down the drain while flushing with large amount of cold water.

12.3 Actual reagent preparation volumes are to be reflected anticipated usage and reagent stability.

13.0 REFERENCES

13.1 United States Environmental Protection Agency, Methods for Chemical Analysis of Water and Waste, Method Number 375.2, August 1993

13.2 American Public Health Association, Standard Methods for the Examination of Water and Wastewater, 21st Edition, Method 4500- SO_4^{2-} G, 2005

13.3 Lachat Instruments, *QuikChem method 10-116-10-2-A, Determination of Sulfate By Flow Injection Analysis Colorimetry*, August 2008

13.4 Lachat Instruments, *Software User Guide*, *Omnion 3.0*, 2004

13.5 Division of Environmental Chemistry, DHMH – Laboratories Administration, *Quality Assurance Plan,* Revision14.0, October 2014

13.6 Division of Environmental Chemistry, DHMH – Laboratories Administration, *Quality Manual,* Revision1.0, November 2014

APPENDICES

Appendix A – Data Review Checklist - Sulfate

Appendix B – Sample Run Log - Sulfate Analysis

APPENDIX **A**

State of Maryland
DHMH - Laboratories Administration
Division of Environmental Chemistry
INORGANICS ANALYTICAL LABORATORY

Data Review Checklist – Sulfate

EPA Method 375.2; revision 2.0

Lab Numbers[1]: _____

Date Collected: _____ Date Analyzed: _____ Analyst: _____

Procedure	Acceptance Criteria	Status*	Comments
Holding Time	28 days @ 4 °C		
Calibration Curve	Corr. Coeff. $\geq$ 0.9950		
External QC[2]	Beginning and end of each run		
	Within acceptable range		
Reagent Blank	< Reporting level (10 mg/L)		
Blank Spike	1 per batch		
	Recovery = 90 – 110%		
Duplicates / Replicates	Every 10th and the last sample or 1/batch, if less than 10 samples		
	RPD $\leq$ 10%		
Matrix Spike	Every 10th and the last sample or 1/batch, if less than 10 samples		
	Recovery = 90 – 110%		
Check Standard	After every 10th sample and at the end of the run		
	Concentration within 90 to 110% of the true value		
Decimal Places Reported	3		
Measured Values	Within calibration range (10 to 100 mg/L)		
Diluted Samples	Correct final calculations		
Changes/Notes	Clearly stated		

* Check (✓) if criteria are met. [1]Include beginning and ending numbers, account for gaps by bracketing numbers.

_____ _____
Analyst's Signature & Date Reviewer's Signature & Date

_____
Supervisor's Signature & Date

[2]QC Sample: _____ Tracking ID: _____

True Value: _____ Acceptable Range: _____

APPENDIX **B**

DHMH Laboratories Administration
Division of Environmental Chemistry
GENERAL CHEMISTRY SECTION
Sample Run Log – Sulfate Analysis
EPA Method 375.2; revision 2.0

Date Analyzed: _____ Analyst: _____

Cup #	Sample ID	Dilution	Concn. (ppm)
Standard Rack 1	Blank		
9	Cal 1, 100 ppm		
10	Cal 2, 80 ppm		
11	Cal 3, 60 ppm		
12	Cal 4, 40 ppm		
13	Cal 5, 20 ppm		
14	Cal 6, 10 ppm		
15	Cal 7, 0 ppm		
Sample Rack 1			
2			
3			
4			
5			
6			
7			
8			
9			
10			

Cup #	Sample ID	Dilution	Conc., ppm
11			
12			
13			
14			
15			
16			
17			
18			
19			
20			
21			
22			
23			
24			
25			
26			
27			

Sample Name	Prep Log ID
Cal 1, 100 ppm	
Cal 2, 80 ppm	
Cal 3, 60 ppm	
Cal 4, 40 ppm	
Cal 5, 20 ppm	
Cal 6, 10 ppm	
Cal 7, 0 ppm	
QC	
Carrier, 0.30 ppm	
Barium chloride	
Barium-MTB	
NaOH, 50%	
NaOH, 0.18N	
HCl, 0.1N	

Lab #	Average	RPD	% Spiked Rec

DETERMINATION OF TOTAL SUSPENDED SOLIDS

Standard Method 2540 D

1.0 SCOPE AND APPLICATION

1.1 The total suspended solids (TSS) are present in sanitary wastewater and many types of industrial wastewater. There are also nonpoint sources of suspended solids, such as soil erosion from agricultural and construction sites.

1.2 TSS is the portion retained on a filer of 1.5 μm (or smaller) nominal pore size.

1.3 This method is suitable for the determination of solids in potable, surface and saline waters, as well as domestic and industrial wastewaters in the range up to 20,000 mg/L. This laboratory reports all values greater than 1 mg/L.

2.0 SUMMARY OF METHOD

A well-mixed sample is filtered through a pre-washed and weighed microfiber filter with 1.5 μm pore size and the residue retained is then dried to a constant weight at 103 to 105 ^{o}C. The increase in weight of the filter represents the total suspended solids.

3.0 INTERFERENCES

3.1 Samples high in dissolved solids, such as saline water, brines and some wastes, may be subject to a positive interference. For such samples, the filter with sample should be washed thoroughly to ensure removal of dissolved solids from the filter.

3.2 Samples with large floating particles, submerged agglomerates of non-homogeneous materials or excessive residue on the filter may form a water-entrapping crust; limit the sample size to that yielding no more than 200 mg residues on the filter pad.

3.3 Samples high in oil and grease may be difficult to dry the residue to a constant weight in a reasonable amount of time.

4.0 HEALTH AND SAFETY

4.1 There is no apparent safety hazard associated with this analysis. However, it is advisable to wear disposable gloves and protective laboratory clothing when handling the samples and to wear autoclave gloves when taking metal trays in or out of the oven.

4.2 Each employee is issued a ***Laboratory Safety Manual*** and is responsible for adhering to the recommendations contained therein.

4.4 A reference file of material safety data sheet (MSDS) is available in lab.

5.0 EQUIPMENT AND SUPPLIES

5.1 Equipment

5.1.1 Balance – Analytical, XS 204, Mettler-Toledo

5.1.2 Computer and printer

5.1.3 Balance Data Transfer Software – *LabX direct balance*, V1.2, Mettler-Toledo

5.1.4 Adapter Cable – USB-RS232, Part # 11103691, Mettle-Toledo

5.1.5 Oven – Isotemp 500 series, 20 to 220^{o}C range, Fisher

5.1.6 Desiccator Cabinet – Stainless steel, cat # 08-645-11, Fisher

5.1.7 Desiccator – Glass with porcelain plate, cat # 08-615B, Fisher

5.1.8 Büchner flask, also known as a vacuum flask, 1000 ml, cat # 10-180F, Fisher

5.1.9 Filter Assembly for using commercially prepared pre-wash and pre-weigh filters

5.1.9.1 Filters – ProWeigh 47 mm glass fiber filters for Total suspended Solids, 1.5 µm pore size, cat # F93447MM-X, Environmental Express

5.1.9.2 Filter Holder – 47 mm polysulfone holder with funnel and base, cat # D0047P, Environmental Express

5.2 Supplies

5.2.1 Desiccants – Silica gel beads, cat # 08-594-17C, Fisher gel

5.2.2 Gloves – Autoclave, cat # 19-013-586, Fisher

5.2.3 Tongs – Stainless steel, cat # 15-186, Fisher

5.2.4 Thermometer – Oven, certified traceable, 20 to 130 ^{o}C, cat # 15-171-5, Fisher

5.2.5 Trays – Stainless steel, cat # 13-361C, Fisher

6.0 REAGENTS AND STANDARDS

6.1 Deionized water

6.2 Quality control (QC) samples

6.3 QC SLD Solid Standards in Water, Inorganic Ventures.

6.4 Universal Solids Standard – Item # 2781, Environmental Express

7.0 SAMPLE COLLECTION, PRESERVATION, AND STORAGE

7.1 Non-representative matter such as large floating particles or submerged agglomerates of non-homogeneous materials should be excluded from the sample if it is determined that their inclusions are not desired in the final result.

7.2 Samples are collected in polyethylene cubitainer,

7.3 Refrigeration or icing at 4 ^{o}C prior to analysis is required to minimize microbiological decomposition of solids. The holding time is 7 days at 4 ^{o}C. However, it is recommended to begin the analysis as soon as possible.

8.0 QUALITY CONTROL

8.1 An initial Demonstration of Performance (IDP) to establish the instrument's capability is required prior to its use in analyzing client's samples. The linearity of calibration range (LCR) and the ability to quantify correctly Quality Control Samples are used to assess performance.

8.2 Deionized water is run as the blank control.

8.3 Replicates are performed on every tenth sample or one replicate per run.

8.4 A QC sample is run quarterly.

8.5 Data acceptance criteria are listed on data review checklist. (Appendix A)

8.6 Balance is professionally serviced and calibrated yearly and is checked with certified external weights and recorded daily.

8.7 A method detection limit (MDL) study is performed once a year by analyzing seven replicates of a low concentration standard in one run. MDL is calculated as follows:

MDL = (t) x (S) where, t = Student's t value for a 99% confidence level and a standard deviation estimate with n-1 degrees of freedom [t = 3.14 for seven replicates] and S = standard deviation of the replicate analyses.
Each new analyst also performs an MDL study or when any changes in the analytical procedure or instrument are made

9.0 PROCEDURE

9.1 Prepare a sample run log sheet (Appendix B) starting with a deionized water blank, replicates for every tenth samples or one per batch, and a QC when needed using the log. StarLIMS samples are recorded on the same run log.

9.2 Sample Analysis

9.2.1 Arrange the run log by Recording the identification number and weight of the ProWeigh filters that is going to be used for each sample. The dish/filter ID is listed in **Dish No**. column, record date of analysis under the **Date column** and enter the weight under **Initial Weight** column (appendix B).

9.2.2 Thoroughly mix sample by inversion. Use about 300ml for Potomac Boat samples and entire (supplied) volume for storm samples. Use a measuring cylinder to measure exact volume of sample filtered. Record the total volume filtered in the run log.

9.2.3 Using forceps, carefully place filter in the filter vacuum assembly. Squirt some distilled water in the filter to wet the filter paper and then turn on the vacuum pump.

9.2.4 Dispense measured volume in the corresponding filter as per your run log.

9.2.5 Rinse the graduated cylinder, filter, non-filterable residue and crucible wall with three successive 10-mL volumes of deionized water and add to the filter and continue suction for about 3 minutes after filtration is completed.

9.2.6 After all liquid has passed through the filter, first disconnect the suction flask from the pump tubing to release the pressure. Then turn off the vacuum. Remove filter and place it back in the original metal pan.

9.2.7 Dry at 103 ^{o} to 105 ^{o}C overnight, cool in a desiccator for 2 hours and determine the 1st final weight.

9.2.8 Turn on the computer. Click on the "**LabX direct balance**" icon.

9.2.9 Click to open TSS folder, select TSS template and enter the sample list. Click file and save the new file name by entering "**yy-mo-day**".

9.2.10 Check the balance with minimum three weights and record in the log book.

9.2.11 After determining the 1st final weight, repeat the cycle of drying, cooling, desiccating and weighing until a constant weight is obtained or until the weight change is less than 4% of the previous weight or 0.5 mg.

9.2.12 Return the filters into the oven for at least one hour, cool in desiccators for two hours, and determine the 2nd final weight.

10.0 DATA ANALYSIS AND CALCULATIONS

10.1 Sample result in the Microsoft Excel table (Appendix B) is formulated according to the following equation:

$$\text{TSS, ppm} = \frac{(\text{wt. of filter \& residue, g} - \text{wt. of filter, g}) \times 1000}{\text{Vol. of sample, mL}}$$

10.2 Calculate the % relative percent difference for the duplicated samples as follows:

$$\text{RPD} = \frac{\text{difference of the duplicates}}{\text{average of the duplicates}} \times 100$$

10.3 The detection limit for this method is 1 ppm.

11.0 DATA AND RECORDS MANAGEMENT

11.1 Instrument Maintenance, external QC and Ongoing Precision and Recovery, are kept in binders and test results are kept in the file cabinet.

11.2 Normal turnaround time for the analysis of samples submitted to this lab will be 2 to 10 days from receipt. Results are reported either in writing on a sample analysis request form or in a print out generated by StarLIMS.

11.3 The Division shall retain all pertinent laboratory records for each sample for a period of five years. At the conclusion of this period, clients shall be given the option to take custody of their sample records. The Division shall not be responsible for retrieving, copying, or transferring laboratory records exceeding the five years custody period.

12.0 WASTE MANAGEMENT

Samples are poured down the drain while flushing with large amount of cold water. Filters are disposed of as regular trash.

13.0 REFERENCES

13.2 United States Environmental Protection Agency, *Methods for Chemical Analysis of Water and Waste,* Method Number 160.2, August, 1993

13.3 American Public Health Association, *Standard Methods for the Examination of Water and Wastewater,* Method Number 2540 D, 21st Edition, 2005

13.4 Division of Environmental Chemistry, Maryland Department of Health and Mental Hygiene, *Quality Assurance Plan,* Revision 14.0, October 2014.

13.5 Division of Environmental Chemistry, Maryland Department of Health and Mental Hygiene, *Quality Manual,* Revision 1.0, November 2014.

APPENDIX A
Division of Environmental Chemistry
INORGANICS ANALYTICAL LABORATORY

Data Review Checklist – Total Suspended Solids (TSS)

Standard Method 2540 D

Lab Numbers[1]:_____

Date Collected: _____ Date Analyzed: _____ Analyst: _____

Procedure	Acceptance Criteria	Status*	Comments
Holding Time	7 days @ 4°C		
Samples Analysis	Started within 5 working days		
Reagent Blank	< 1 mg/L		
Duplicates/Replicates	A minimum of 10% of the samples or 1/batch, if less than 10 samples		
	RPD ≤ 15%		
External QC[2] Analyze quarterly	Within acceptable range		
	Last date analyzed:		
Decimal Places Reported	DNR split samples: 3		
	All other DNR samples a. Results < 1 mg/L: 1 decimal place; report with "L" letter b. Results ≥ 1 mg/L: 0 decimal place		
	All samples on multiple sheets: 0		
Calculations	Done correctly		
Changes/Notes	Clearly stated		

* Check (√) if criteria are met. [1]Include beginning and ending numbers, account for gaps by bracketing

_____ _____
Analyst's Signature & Date Reviewer's Signature & Date

_____
Supervisor's Signature & Date

[2]External QC
Identification = _____
True Value = _____ ppm
Range = _____ ppm

APPENDIX B

DHMH – Laboratories Administration
Division of Environmental Chemistry
INORGANICS ANALYTICAL LABORATORY

Sample Run Log – Total Suspended Solids (TSS)

Standard Method 2540 D

Analyst: _____

Lab No.	Vol. Filtered (L)	Dish No	Initial Wt Date	Initial Wt (gm)	Final Wt (1) Date	Final Wt (1) (gm)	Final Wt (2) Date	Final Wt. (2) (gm)	Net Wt (gm)	TSS (mg/L)	Average & RPD

STANDARD OPERATING PROCEDURE

DETERMINATION OF 5 - Day Biochemical Oxygen Demand

Standard Method 5210 B

1.0 SCOPE AND APPLICATION

1.1 The biochemical oxygen demand (BOD) test is used for determining the relative oxygen requirement of wastewaters, effluents, polluted waters, and streams. The test has its widest application in measuring waste loadings to treatment plants and in evaluating the BOD-removal efficiency of such treatment system. The application of the test to organic waste discharges allows calculation of the effect of the discharges on the oxygen resources of the receiving water.

1.2 The BOD determination is an empirical test which measures the dissolved oxygen utilized during a specified incubation period for the biochemical degradation of organic material (carbonaceous demand) and the oxygen used to oxidize inorganic material such as sulfides and ferrous irons. It also may measure the amount of oxygen used to oxidize reduced forms of nitrogen (nitrogenous demand) unless their oxidation is prevented by an inhibitor. The standard test conditions include dark incubation at 20 ^{o}C for five days. The actual environmental conditions of temperature, biological population, water movement, sunlight, and oxygen concentration cannot be actually reproduced in the laboratory. Results obtained must take into account the above factors when relating BOD results to stream oxygen demands.

2.0 SUMMARY OF METHOD

2.1 Appropriate dilutions of each sample and the quality control samples are incubated for 5 days (BOD_5) at 20 ^{o}C in the dark. The reduction in dissolved oxygen concentration during the incubation period yields a measure of the biochemical oxygen demand.

3.0 INTERFERENCES

3.1 Residual chorine can interfere in this determination and it is neutralized with Na_2SO_3, if present. Hach's USEPA-accepted DPD (N, N-diethyl-p-phenylenediamine) colorimetric method is used to detect any free chlorine in the sample.

3.2 The source water used for BOD sample dilution must be free of heavy metals, specifically copper, and toxic substances such as chlorine that can interfere with BOD measurements. Protect source water quality by using clean glassware, tubing, and bottles. Storage of prepared dilution water for more than 24 h after adding nutrients, minerals, and buffer is not recommended unless dilution water blanks consistently meet quality control limits.

3.3 Oxidation of reduced forms of nitrogen, mediated by micro-organisms, has been considered interference in the determination of BOD and can be prevented by an inhibitory chemical and reported results as carbonaceous biochemical oxygen demand (CBOD).

3.4 Exclude all light during the 5 day incubation period to prevent the possibility of photosynthetic production of dissolved oxygen (DO).

4.0 HEALTH AND SAFETY

4.1 Good laboratory practices should be followed during reagent preparation and instrument operation.

4.2 Each employee is issued a ***Laboratory Safety Manual*** and is responsible for adhering to the recommendations contained therein.

4.3 Use absorbent towels if material is spilled and wash residual into drain.

4.4 Each chemical should be regarded as a potential health hazard. A reference file of material safety data sheet (MSDS) is available in lab.

5.0 EQUIPMENT AND SUPPLIES

5.1 Equipment

5.1.1 YSI Model 5100 dissolved oxygen meter

5.1.1.1 Dissolved oxygen (DO) probe

5.1.1.2 Membrane replacement kits for DO probe

5.1.1.3 Mantech AutoMax 122 Autosampler with pumps

5.1.1.4 Computer and printer

5.1.2 Incubation room, thermostatically controlled at $20 \pm 1^{o}C$

5.1.3 pH meter – Accumet pH meter 15, Fisher Scientific

5.1.4 Magnetic stirrer

5.1.5 Buret – 50 mL

5.1.6 Drying oven – isotemp, gravity flow convection, 103 ^{o}C to 105 ^{o}C

5.1.7 Air compressor – 135 psi, Westward

5.2 Supplies

5.2.1 BOD bottles – 300 mL disposable bottles (cat. # D1001), botttle stoppers (cat. # D1025), and overcaps (cat. # D1050), Environmental Express

5.2.2 Carboy with spigot – 20 L capacity

5.2.3 Graduated Cylinders – 25, 50, 100, and 250 mL

5.2.4 Micropipetter – adjustable volume ranges from 1.0 to 5.0 mL

5.2.5 Pipet tips – 5000 µL

5.2.7 Plastic beakers – polypropylene, 1000 mL,

5.2.8 Membrane kit for BOD probe – cat. # 5906, YSI

5.2.9 Filter Unit, 0.45 µm – Nalgene disposable sterilization filter unit, cat. # 09-740-25B, Fisher

5.2.10 Tubes – polypropylene with snap caps, sterile, 14 mL, cat. # 14-959B, Fisher

5.2.11 Glass pipettes – volumetric, class A, 5 mL

5.2.12 Flasks – volumetric, class A, 500 mL and 1000 mL

5.2.13 Glass rods

5.2.14 Stirring bars

5.2.15 Weighing pans – aluminum, cat. #D57-144, Labsources, Inc.

6.0 REAGENTS

6.1 Dilution water

6.1.1 Aerate 19 liters (5 gallons) of deionized water in a 20 L carboy in the 20 ^{o}C room for 30 minutes. The dissolved oxygen concentration of water used for BOD test must be at least 7.5 mg/L. Following aeration, leave carboy to sit overnight in 20 ^{o}C room with the cap loosened to allow water to equilibrate.

6.1.2 Empty one premixed pillow of BOD Nutrient Buffer (Hach cat. # 14863-98) into aerated water (6.1.1) at 20 ^{o}C. Mix well. Prepare dilution water one hour before use.

6.2 Glucose-Glutamic acid (GGA) solution

6.2.1 Dry few grams each of glucose or dextrose and glutamic acid in aluminum weigh pans for 1 hour at 103 ^{o}C. Cool to room temperature in a dessicator.

6.2.2 Weigh out 0.15 g each of dextrose and glutamic acid and dissolve in 800 mL of deionized water in a 1 L volumetric flask. Dilute to mark and mix well. Prepare fresh immediately before use.

6.2.3 Instead of preparing fresh GGA solution each time, the solution prepared in 6.2.2 can be sterilized by filtering through a disposable sterilization filter unit, divided and stored in small volumes. If this procedure is followed, pour about 12 mL aliquots into each sterile 14 mL polystyrene

tube, snap cap back on the tube, label, and store in the refrigerator. Prepare every two months.

6.2.4 Premade GGA is also available (Man-Tech Cat. No P17801). To prepare the standards, simply add the content of the 6 mL vial into each of the two BOD bottles marked for GGA.

6.3 Seeding material, prepare daily

6.3.1 One bottle of wastewater from the Cox Creek Wastewater Treatment Plant is delivered to the laboratory every Tuesday. Store the wastewater in the incubation room.

6.3.2 Pour the supernatant into an Erlenmeyer flask about an hour before beginning the run to allow solids to settle to the bottom of the flask. The amount of supernatant to be added to each BOD bottle is between 1.5 mL to 3.0 mL depending on the color, odor and density of the wastewater.

6.4 Sample pH

6.4.1 Calibration buffers – pH 4.0, pH 7.0, and pH 10.0 - Fisher cat. # SB105,

6.4.2 Sulfuric acid (H_2SO_4), 1M – Slowly and while stirring, add 2.8 mL of conc. H_2SO_4 to 80 mL of deionized water. Dilute to 100 mL. Mix well, label and store.

6.4.3 Sodium hydroxide (NaOH), 1N – Dissolve 4 g of NaOH in 80 mL of deionized water. Dilute to 100 mL.

6.5 Dechlorination

6.5.1 DPD free chlorine reagent power – cat. # 14070-99, Hach

6.5.2 Starch soluble for iodometry – cat. # 516-100, Fisher

6.5.3 Sodium sulfite solution (Na_2SO_3) – Dissolve 0.157 g of Na_2SO_3 in 100 mL of deionized water. This solution is not stable; prepare fresh daily.

6.5.4 Potassium iodide (KI) solution – Dissolve 10 g of KI in 100 mL deionized water. Mix well.

6.5.5 Acetic acid (CH_3COOH), 1:1 – Mix 20 mL deionized water with 20 mL glacial acetic acid.

6.5.6 Nitrification inhibitor – 2-chloro-6-(trichloro methyl) pyridine (TCMP), cat. # 2533, Hach

6.5.7 External Quality Control Sample – QC-DEM-WP, Spex Certiprep Inc.

7.0 SAMPLE COLLECTION, PRESERVATION, AND STORAGE

Keep samples at or below 4 ^{o}C from the time of collection and analyze within 48 hours after collection.

8.0 QUALITY CONTROL

8.1 Dilution water quality check: The dilution water blank, prepared in 6.1, serves as a check on quality of unseeded dilution water and cleanliness of incubation bottles. The DO uptake in 5 days must not be more than 0.20 mg/L. If this value exceeds 0.20 mg/L, then evaluate the cause and make appropriate corrections.

8.2 Glucose-glutamic acid check: The glucose-glutamic acid check solution is the primary basis for establishing precision and accuracy and is the principal measure of seed quality and analytical technique. For the 300 mg/L mixed primary standard, the average 5 days BOD must fall within the range of 198 ± 30.5 mg/L. If the average value falls outside this range, evaluate the reason and take appropriate actions. Consistently high values can indicate the use of too much seed suspension, contaminated dilution water, or the occurrence of nitrification. Consistently low values can indicate poor seed quality, use of insufficient quantity of seed suspension, or the presence of toxic materials. If low values persist, prepare a new mixture of glucose and glutamic acid and check the sources of dilution water and the seed.

8.3 Minimum residual DO and minimum DO depletion*:* Only the dilutions resulting in a DO depletion of at least 2.0 mg/L and a residual DO of at least 1.0 mg/L after 5 days of incubation are considered to produce valid data.

8.4 Seed Control: The DO uptake attributable to the seed should be between 0.6 -1.0 mg/L. The volume of seed added should be adjusted in order to meet the required range of 198 ± 30.5 mg/L for glucose-glutamic acid check.

8.5 An external quality control sample with a known BOD value is analyzed each quarter.

8.6 The YSI dissolved oxygen meter is calibrated in air (water saturated), i.e. the probe is parked in a BOD bottle containing 1" of water.

8.7 Data acceptance criteria are listed in the data review checklist (Appendix A).

8.8 Laboratory participates in ERA WatR Pollution (WP) Proficiency Testing annually.

9.0 PROCEDURE

9.1 Sample preparation:

9.1.1 Prepare the sample run list for checking color, odor, pH and chlorine and for dilutions. (Appendix B)

9.2 Check samples for residual chlorine.

9.2.1 Using the Hach Swiftest dispenser, insert DPD free chlorine reagent powder into each test tube, add about 10 mL of sample and observe for any color

change occurring within a few seconds. A pink color indicates presence of chlorine and therefore the samples(s) must be dechlorinated.

9.2.2 Determine the required volume of Na2SO3 needed to dechlorinate on a 50 mL portion of the pH adjusted sample. Add 0.5 mL of 1:1 acetic acid (6.5.5), 0.5 mL of KI solution (6.5.4) and a few drops of starch solution to sample. Using a 50 mL buret, titrate with Na2SO3 (6.5.3) solution to the starch-iodine (blue) end point. Record the volume used. Calculate and add the required volume of Na2SO3 solution to the pH adjusted portion of the sample (9.3.3).

9.3 Check sample pH

9.3.1 Label 1 L polypropylene beakers with the sample numbers. Pour about 500 mL of samples into 1 L beakers. Pour 100 mL of sample if it has strong sewage odor.

9.3.2 Standardize the pH meter using pH 4, 7 and 10 buffers. Tap the pH meter screen twice to access the menu. Tap pH and then tap "STD". Touch "CLEAR" to delete previous buffers. Follow the screen prompts to standardize the new buffers. Record the results in the pH meter log. To read sample pH: Touch "MEASURE". Read pH of all the samples making sure they are stirred during the measurement. Leave the pH meter on standby when finished. The pH of the samples out of the range of 6.5 to 7.5 must be adjusted to this range.

9.3.3 Adjust each sample to pH 6.5 to 7.5 with 1N NaOH or 1M H_2SO_4 and record the final pH.

9.4 Sample dilution:

9.4.1 Bring samples to BOD room temperature (20 ^{o}C) before making dilutions.

9.4.2 Check samples for color and odor.

9.4.3 Dilutions are prepared directly in BOD bottles. Transfer 200 and 100 mL aliquots of each prepared stream sample, 50, 25, 10 and 5 mL aliquots of each prepared sewage sample, and 10, 5, 1 and 0.5 mL aliquots of each prepared strong industrial wastes, as appropriate, into labeled BOD bottles using class A graduated cylinders and volumetric pipets. Rinse the cylinder between samples. Dilutions may need to be adjusted to reflect the qualities of the sample. Place the bottles in the correct order in the rack.

9.5 Nitrification inhibition:

9.5.1 If nitrification inhibition is desired add 3 mg of TCMP (6.5.6) to each 300 mL bottle before capping.

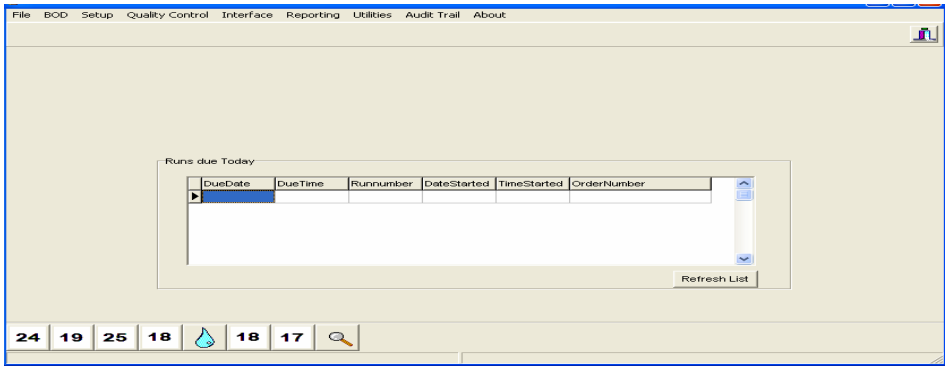
9.5.2 Note the use of nitrification inhibition in the reporting results.

9.6 Prepare autosampler using the "PC-BOD" software:

9.6.1 Warm up YSI 5100 for at least 30 minutes. Ensure that it is set in **REMOTE** mode.

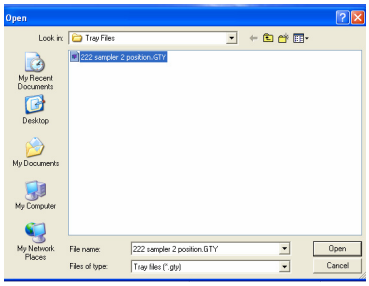
9.6.2 On the computer desktop locate the software icon.

Double click on the icon and the software will open to the main screen. If the icon is not present, open the software by clicking on the desktop 'Start' menu, followed by 'All Programs' and select 'PC-BOD'.



9.6.3 Under **BOD** select **MANUAL.**

9.6.3.1 Click on the '**Load Tray from Folder**' button. The window shown at right will appear.

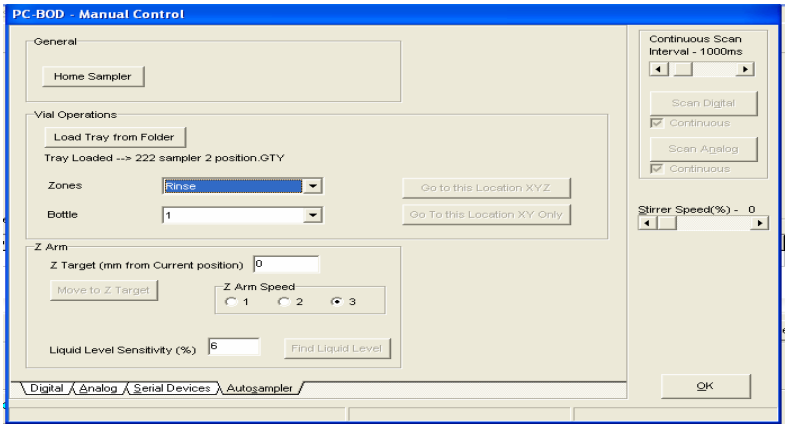


9.6.3.2 Click on the tray file named *271 sampler…* and then click on the '**Open**' button.

9.6.3.3 The'**Home Sampler**' button will become active and the 'zones' and 'bottle' windows will be filled in.

9.6.3.4 Click on the '**Home Sampler**' button. The sampler will move to the home position and the buttons to the right of'**zones**' & **'bottles'** will become active. If the sampler is already in the home position, it will appear that nothing is happening but within a few seconds the two buttons will become active. Remove the rack and place a waste beaker in bottle position 3.

9.6.3.5 To move the autosampler to a specific location first select the following zone: -Bottle: allows DO probe to go into a bottle



9.6.3.6 Select the bottle location to move to by using the drop down menu. For example selecting 'Bottle' and '3' will allow the DO probe to go to the 3rd bottle position.

9.6.3.7 To move the autosampler to the specified location click on **'Go to this location XY only'** to move above the bottle position.

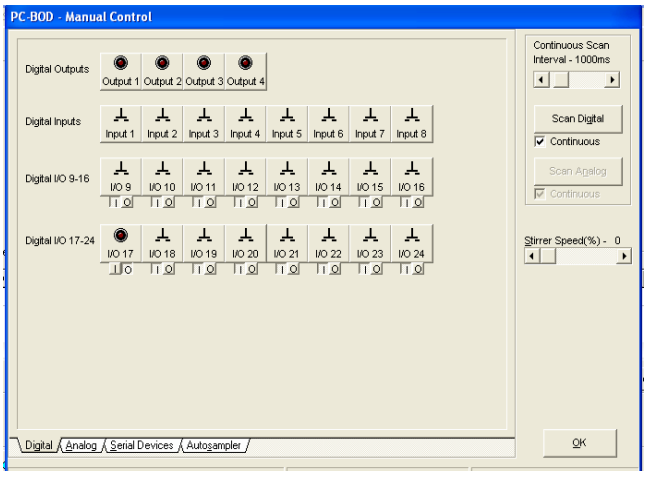
9.6.3.8 To move the autosampler only in the Z direction (up and down), enter the number of millimeters to move in the box next to '**Z Target (mm from current position)**'.Use a '–' sign before the number of millimeters to indicate moving in a downward direction. Click '**Move to Z Target'** to move the sampler. For example, entering -43 will move the Z arm down 43mm.

9.6.3.9 Move the autosampler to bottle location **3** and lower it into a waste collection beaker by moving the autosampler down the Z axis by three **-43mm** increments.

9.6.4.0 Prime the pumps with the seed, nutrient water and rinse water.

9.6.4.1 Open '**Digital**' Tab: This tab allows the pumps and stirrer to be turned on or off. Click on the button listed below to turn the device on or off.

Output 1 – dilution pump
Output 2 – seed pump
Output 3 – inhibitor pump
Output 4 – DO probe stirrer
Output 17 – rinse pump



9.6.4.2 Turn on the seed pump,(**Output2**), ensuring that all the rinse water left in the line has been emptied into the waste beaker and the seed is being drawn completely through the line. Turn it off when the seed is dripping into the waste beaker.

9.6.4.3 Turn on the dilution pump, (**Output 1**), ensuring that all the rinse water left in the line has been emptied into the waste beaker and the nutrient water is being drawn through the line. Turn it off.

9.6.4.4 Return the Autosampler to the home position.
1.Open the **Autosampler** tab.
2.Click on '**home sampler'**
3.Click '**OK**'.

9.7 Choose Calibration Schedules and Method.

9.7.1 From the run screen (**BOD/Run BOD**), click on the '**Calibration Schedule' and** choose '**YSI1500 Barometer Cal**'from the drop down menu.

9.7.2 Select the method to use by clicking on the **'Schedule'** button. Choose pump schedule '**3PUMPSJBF**'

9.8 Setting up a run manually.

9.8.1 From the run screen (**BOD/Run BOD**), click on the '**Edit'** followed by the '**Add X Rows**' button. Enter the number of rows that need to be added to the one already on the grid to give one row per bottle in the run. Click '**OK**' and the rows will be added.

9.8.2 Build the batch starting with one stabilizing water blank, two duplicated water blanks, three seeds at 10, 15 and 20 mL, two duplicated mixtures of 5 mL of G/G with 2-3 mL of seeds followed by 2 to 5 different dilutions of each sample plus 2-3 mL of seeds. See appendix C for an example run and enter as shown.

9.8.3 Fill in the columns on the template. To remove extra lines, click the '**Delete Highlighted Sample**' button. Do not leave blank lines in the template.

9.8.4 Click '**Done Edit**' and the batch will be set up.

9.8.5 Click the '**Auto-Generate Order Number'** button. Enter operator's initials in the box in the upper left corner of the screen.

9.8.6 Load marked bottles into the autosampler racks.

9.8.7 Place the rack containing the first samples onto the autosampler.

9.8.8 Press the '**Start**' button to begin calibration and sample analysis.
When prompted enter the rack number currently on the autosampler and press '**OK**'.

9.8.9 Following the screen prompts with regard to calibrating the autosampler and recording the results in the book.

9.8.10 Continue following the screen prompts to allow the auto dilutor to seed, dilute and take an initial D.O. reading of all the samples in the rack. If there are multiple racks the program will prompt for insertion of them at the correct time.

9.9 When a sample is supersaturated.

9.9.1 Stop, delete initial DO readings higher than 9.2.

9.9.2 Shake the diluted, seeded sample in the designated container to remove excess DO

9.9.3 Restart the run and the autosampler will begin with the first sample without a reading.

9.10 Incubation: After all the samples in a rack have been diluted, seeded and had an initial DO reading taken, remove the rack from the autosampler. Place a stopper and cap on each bottle before incubating the sealed bottles for 5 days in the 20^{o}C incubation room with the lights turned off.

9.11 Read final DO:

9.11.1 Turn on the YSI 1500 and allow to warm up for 30 minutes. Press the **Mode**' button then choose '**Remote**' from among the options.

9.11.2 Loading an Existing Run in the computer.

9.11.2.1 Open the '**PC BOD'** program.

9.11.2.2 On the main screen click on '**BOD**' and then select '**Run BOD**'

9.11.2.3 Choose the '**Load Existing Runs**' tab. On this screen there are 4 buttons which indicate runs in various stages of completion. Choose '**Finals Due Today'** and highlight the row containing the appropriate run.

9.11.2.4 Click on '**Load Selected**' Enter the operators initials in upper right window.

9.11.2.5 Place the rack with the samples to be run onto the autosampler.

9.11.2.6 To begin the run click on the '**Start**' button and enter the number of the rack currently on the autosampler when prompted.

9.11.2.7 Follow the program prompts to calibrate the probe and record the readings in the log book.

9.11.2.8 Continue following the program prompts to take the final D.O. readings.

9.12 Monthly maintenance of BOD probe

9.12.1 Prepare the oxygen probe electrolyte by filling the bottle included with the kit to neck with deionized water. Shake well until crystals are dissolved.

9.12.2 Remove the old membrane cap assembly from the probe. Wipe clean the metal tip of the probe.

9.12.3 Take a new membrane cap assembly and fill in with the fresh electrolyte solution and then screw the cap assembly onto the probe.

9.12.4 Always park the probe in a BOD bottle containing one inch of D.I. water when not in use.

10.0 DATA ANALYSIS AND CALCULATIONS

10.1 All the calculations are preformed automatically by BOD Analyst software using the following equations:

10.1.1 Amount of dissolved oxygen consumed during the incubation period:

O_2 Depletion (mg/L) = Initial DO – Final DO

10.1.2 Seed factor used for correcting the BOD test for oxygen depletion resulting from the presence of seed:

$$\text{Seed Factor (mg/L)} = \frac{O_2 \text{ Depl in seed control}}{\text{Vol seed in seed control}} \text{ x Vol seed in sample}$$

10.1.3 BOD of the samples:

$$\text{BOD (mg/L)} = \frac{O_2 \text{ Depl in sample} - \text{Seed Factor}}{\text{Sample Volume, ml}} \text{ x Bottle Volume, mL}$$

10.2 If more than one sample dilution meets the acceptance criteria, report the average calculated by the software program.

10.3 If the O_2 depletion is less than 2 mg/L with 200 mL portion (maximum sample volume) of the sample, report the result from this dilution.

10.4 If all the sample dilutions produce a final DO of less than 1.0 mg/L, report the result from the highest dilution with a > sign.

11.0 DATA AND RECORDS MANAGEMENT

11.1 All Quality Control data are kept in a binder labeled as “Quarterly QC for BOD”.

11.2 Normal turnaround time for BOD samples submitted to this lab is 7 to 10 days from receipt with a sample holding time of 2 days. Results are reported in writing on a sample analysis request form.

11.3 The Division shall retain all pertinent laboratory records for each sample for a

period of five years. At the conclusion of this period, clients shall be given the option to take custody of their sample records. The Division shall not be responsible for retrieving, copying, or transferring laboratory records exceeding the five years custody period.

12.0 WASTE MANAGEMENT

12.1 It is laboratory's responsibility to comply with all state, federal and local regulations governing waste management, particularly the hazardous waste identification rules and disposal restrictions, and to protect the air, water and land buy minimizing and controlling all releases from fume and bench operation.

12.2 Samples and standards are poured down the drain while large amount of water is running.

13.0 REFERENCES

13.1 United States Environmental Protection Agency, *Methods for Chemical Analysis of Water and Waste*, Method Number 405.1, August, 1993.

13.2 American Public Health Association, Standard Methods for the Examination of Water and Wastewater, 21st Edition, Method 5210 B, 2005.

13.3 YSI BODANALYST Operations Manual, 1999.

13.4 YSI 5905/5010 BOD Probe Instruction Manual, 1999.

13.5 PC-BOD Operator's Manual – Man Tech 2009

13.6 Division of Environmental Chemistry, Maryland Department of Health and Mental Hygiene, *Quality Assurance Plan,* Revision 14.0, 2014

13.7 Division of Environmental Chemistry, Maryland Department of Health and Mental Hygiene, *Quality Assurance Manual,* Revision 1.0, 2014

APPENDIX A

Division of Environmental Chemistry
INORGANICS ANALYTICAL LABORATORY

Data Review Checklist – BOD_5

Standard Method 5210 B

Lab Numbers[1]: _____

Date Collected: _____ Date Analyzed: _____ Analyst: _____

Procedure	Acceptance Criteria	Status*	Comments
Holding Time	48 hours @ 4 °C		
Chlorine	Neutralized if present		
pH	Between 6.5 to 7.5; adjusted if out of range		
Initial DO	< 9.20 mg/L at 20 °C		
Incubation Period	5 days		
DO uptake of dilution water	< 0.20 mg/L		
DO uptake of seeded dilution water (seed factor)	0.60 to 1.00 mg/L		
BOD_5 for Glucose/Glutamic Acid (G/GA) solution	198 ± 30.5 mg/L		
Sample dilutions	Meet the requirements: Final DO ≥ 1.00 mg/L and DO depletion ≥ 2.00 mg/L		
	Decide on the value to be reported if requirements are not met.		
External QC[2] Analyzed quarterly	Last date analyzed		
	Within acceptable range		
Decimal Places Reported	1		
Reported Values	≥ 2 mg/L; concentrations below this value reported with < sign for Chesapeake Bay samples; as < 2 mg/L for all other samples.		
Changes/Notes	Clearly stated		

[1]Include beginning and ending numbers, account for gaps by bracketing.

_____ _____
Analyst's Signature & Date Reviewer's Signature & Date

_____
Supervisor's Signature & Date

[2]QC Sample: _____ Tracking ID: _____
True Value = _____ Acceptable Range = _____

APPENDIX B

Division of Environmental Chemistry

INORGANICS ANALYTICAL LABORATORY

Sample Run Log –BOD_5

Standard Method 5210 B

Date: _____ Analyst: _____

Lab #	Sample Type	Dilution	Color	Odor	pH	pH Adj. to	Chlorine	Chl. Neutr

Sample Name	Tracking ID
pH 4 Buffer	
pH 7 Buffer	
pH 10 Buffer	
Seeds	

Sample Name	Prep Log ID
H_2SO_4, 1M	
NaOH, 1N	
G/GA	
Dilution water	

APPENDIX C

Division of Environmental Chemistry
INORGANICS ANALYTICAL LABORATORY

EXAMPLE OF BATCH

Bottle#	Sample name	Sample Volume (mL)	Seed Volume(mL)
1	Calib		
2	Blank		
3	Blank		
4	Blank		
5	Seed		10
6	Seed		15
7	Seed		20
8	BOD GGA	5	3
9	BOD GGA	5	3
10	WW 1111	100	3
11	WW 1111	200	3
12	WW E12001111001	50	3
13	WW E12001111001	100	3
14	WW E12001111001	200	3
15			

APPENDIX D

Division of Environmental Chemistry
INORGANICS ANALYTICAL LABORATORY

Troubleshooting

PROBLEM	CAUSE	SOLUTION
Autosampler jam.	Tangled lines.	Straighten the lines. Exit the Run. Home the Sampler. Reload the Run.
D.O. readings inconsistent/ unexpected.	Probe membrane no longer intact.	Change membrane.
Initial Blank readings too high.	Dilution water supersaturated.	Degas carboy with Helium gas for 30 seconds.
Initial Sample readings too high.	Sample is supersaturated.	Stop the Run. Pour diluted sample into a shaker and shake for 30 seconds. Return to BOD bottle and replace in rack. Delete the D.O. reading in EDIT mode. Restart the run.

DETERMINATION OF TOTAL DISSOLVED SOLIDs (TDS)

Standard Method 2540 C

1.0 SCOPE AND APPLICATION

1.1 Waters with high dissolved solids generally are of inferior palatability and may induce an unfavorable physiological reaction in the transient consumer. For these reasons, a limit of 500 mg dissolved solids/L is desirable for drinking waters.

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

1.3 This method is suitable for the determination of solids up to 20,000 mg/L.

2.0 SUMMARY OF METHOD

An aliquot of a well mixed sample is filtered through a Gooch crucible containing a glass fiber filter with 1.5 µm pore size. The filtrate is evaporated to dryness in a weighed dish and dried to constant weight at 180^{o}C. The increase in dish weight represents the total dissolved solids.

3.0 INTERFERENCES

3.1 Highly mineralized waters with a considerable calcium, magnesium, chloride, and/or sulfate content may be hygroscopic and will require prolong drying, desiccation and rapid weighing.

3.2 Sample high in bicarbonate require prolonged drying at 180^{o}C to insure that all the bicarbonate is convert to carbonate.

3.3 Too much residue in the evaporating dish will crust over and entrap water that will not be driven off during drying. Limit sample to no more than 200 mg residue.

4.0 HEALTH AND SAFETY

4.1 There is no apparent safety hazard associated with this analysis. However, it is advisable to wear disposable gloves and protective laboratory clothing when handling the samples and to use tongs and to wear heat resistant gloves when taking dishes in or out of the oven.

4.2 Each employee is issued a ***Laboratory Safety Manual*** and is responsible for adhering to the recommendations contained therein.

4.3 A reference file of material safety data sheet (MSDS) is available in lab.

5.0 EQUIPMENT AND SUPPLIES

5.1 Equipment

5.1.1 Balance – Analytical, XS 204, Mettler-Toledo

5.1.2 Computer and printer

5.1.3 Balance Data Transfer Software – *LabX direct balance*, V1.2, Mettler-Toledo

5.1.4 Adapter Cable – USB-RS232, Part # 11103691, Mettle-Toledo

5.1.5 Oven – Isotemp 500 series, 20 to 220 ^{o}C range, Fisher

5.1.6 Desiccator – Glass with porcelain plate, cat # 08-615B, Fisher

5.1.7 Steam Bath under the hood

5.2 Supplies

5.2.1 Filters – Glass microfiber, 24 mm diameter, 1.5 µm retention, cat # 934-AH, Whatman

5.2.2 Crucibles – Gooch, 40 mL capacity, cat # 08-198E, Fisher

5.2.3 Crucible Holder – Rubber with removable glass stem ground at 45^{o} angle, cat # 08-285, Fisher

5.2.4 Flask – Filtering with tabulation, 1000 ml, cat # 10-180F, Fisher

5.2.5 Desiccants – Silica gel beads, cat # 08-594-17C, Fisher gel

5.2.6 Gloves – Autoclave, cat # 19-013-586, Fisher

5.2.7 Tongs – Stainless steel, cat # 15-186, Fisher

5.2.8 Thermometer – Oven, certified traceable, +20 to +130 ^{o}C, cat # 15-171-5, Fisher

5.2.9 Trays – Stainless steel, cat # 13-361C, Fisher

5.2.10 Evaporating dishes – Pyrex glass, 80 mm diameter, cat # 08-710A, Fisher

5.2.11 Cylinder – Class A, Pyrex certified, 50 mL, cat # 08-553A, Fisher

5.2.12 Cubitainer – Plastic Bottle, commodity # 17568-140000, DHMH Lab Adm Warehouse

6.0 REAGENTS AND STANDARD

6.1 Deionized water

6.2 Quality Control Sample – Order from an accredited source

7.0 SAMPLE COLLECTION, PRESERVATION, AND STORAGE

7.1 Samples are collected in polyethylene cubitainer,

7.2 Refrigeration or icing at 4^{o}C prior to analysis is required to minimize microbiological decomposition of solids. The holding time is 7 days at 4^{o}C.

7.3 Bring samples to room temperature before analysis.

8.0 QUALITY CONTROL

8.1 Deionized water is run as the blank control.

8.2 Replicates are performed on every tenth sample or one replicate per run.

8.3 A QC sample is run quarterly.

8.4 Data acceptance criteria are listed on the data review checklist. (Appendix A)

8.5 Balance is professionally serviced and calibrated once a year and is checked with external weights by user with each run.

8.6 Laboratory participates in ERA WatR Pollution (WP) Proficiency Test.

9.0 PROCEDURE

9.1 Prepare evaporating dishes by heating clean dishes to 180^{o}C for at least one hour, cooling down in the desiccator for 2 hours.

9.2 On the computer, open the TDS template in TDS file and Prepare a sample run log, starting with a deionized water blank (used in the preparation of the QC) and the QC followed by samples. Replicate every tenth sample or one per batch (Appendix B).

9.3 Check the balance with a minimum of three standard weights (20, 50 and 100g) and record the readings in the logbook.

9.4 Weigh the empty dishes and then transfer the weights to the sample table by pressing the Data Transfer Key on the balance. Set the weighed dishes aside until used for evaporation step.

9.5 Sample analysis

9.5.1 Insert the glass-microfiber filter into the bottom of a Gooch crucible with wrinkled side up. Place the crucible on top of a clean suction flask with a Gooch crucible holder.

9.5.2 Apply vacuum and wash filter with three successive 20 mL of deionized water. Continue suction to remove all traces of water. Discard washings.

9.5.3 Shake to mix sample in the cubitainer, then measure 50 mL with a 50 mL class A graduated cylinder.

9.5.4 Pour the measured sample onto the filter with applied vacuum. Wash with 3 successive 10-mL volumes of deionized water, and continue suction, until filtration is complete.

9.5.5 Transfer total filtrate (with washings) to a weighed evaporating dish (9.4) that has been placed on the steam bath.

9.5.6 Gently evaporate to dryness on the steam bath by regulating the flow of steam so that sample does not bump or pop from the dish. Remove the dish when done.

9.5.7 Dry the evaporated sample overnight in an 180^{o}C oven.

9.5.8 Cool in a desiccator for at least 2 hours and read the first final weight.

9.5.9 Put the dish back into the oven for at least 1 hour. Cool in a desiccator for 2 hours and read the second final weight.

9.5.10 Repeat the cycle of drying, cooling, desiccating and weighing until a constant weight is obtained or until the weight change is less than 0.5 mg.

10.0 DATA ANALYSIS AND CALCULATIONS

10.1 Calculate the TDS according to the following equation:

$$\text{Total Dissolved Solids, mg/L} = \frac{\text{wt of dried residuen \& dish, mg - wt of dish, mg}}{\text{vol of sample}} \text{ x } 1000$$

10.2 Calculate the Relative percent Difference for the duplicated samples as follows:

$$RPD = \frac{\text{difference of the duplicates}}{\text{average of the duplicates}} \times 100$$

10.3 The detection limit for this method is 2 ppm.

11.0 DATA AND RECORDS MANAGEMENT

11.1 Instrument maintenance, instrument calibration, and quality control data are kept in binders and test results are kept in the file cabinet.

11.2 Normal turnaround time for samples submitted to this lab will be 2 to 10 days from receipt. Results are reported in writing on a sample analysis request form.

11.3 The Division shall retain all pertinent laboratory records for each sample for a period of five years. At the conclusion of this period, clients shall be given the option to take custody of their sample records. The Division shall not be responsible for retrieving, copying, or transferring laboratory records exceeding the five years custody period. Wastewater Laboratory keeps all original copies of the data records

12.0 WASTE MANAGEMENT

Samples are poured down the drain while flushing with large amount of cold water. Filters are disposed of as regular trash.

13.0 REFERENCES

13.1 United States Environmental Protection Agency, *Methods for Chemical Analysis of Water and Waste,* Method 160.1 August, 1993

13.2 American Public Health Association, *Standard Methods for the Examination of Water and Wastewater ,* Method 2540 C, 21st Edition, 2005

13.3 Division of Environmental Chemistry, Maryland Department of Health and Mental Hygiene, *Quality Assurance Plan,* Revision 14.0, 2014

13.4 Division of Environmental Chemistry, Maryland Department of Health and Mental Hygiene, *Quality Assurance Manual,* Revision 1.0, 2014

APPENDIX A

DHMH – Laboratories Administration
Division of Environmental Chemistry
INORGANICS ANALYTICAL LABORATORY

Data Review Checklist – Total Dissolved Solids (TDS)

Standard Method 2540 C

Lab Numbers[1]:_____

Date Collected: _____ Date Analyzed: _____ Analyst: _____

Procedure	Acceptance Criteria	Status*	Comments
Holding Time	7 days @ 4°C		
Sample Analysis	Started within 5 working days		
Reagent Blank	< 2 mg/L		
Duplicates/Replicates	A minimum of 10% of the samples or 1/batch, if less than 10 samples		
	RPD ≤ 10%		
External QC[2]	Analyzed with each set of samples		
	Within acceptable range		
Decimal Places Reported	0		
Reporting Level	2 mg/L; concentrations below this value reported as < 2 mg/L		
Calculations	Done correctly		
Changes/Notes	Clearly stated		

*Check (√) if criteria are met. [1]Include beginning and ending numbers, account for gaps by bracketing.

_____ _____
Analyst's Signature & Date Reviewer's Signature & Date

_____
Supervisor's Signature & Date

[2]QC Sample: _____ Tracking ID: _____

True Value = _____ Acceptable Range = _____

APPENDIX B

DHMH – Laboratories Administration
Division of Environmental Chemistry
INORGANICS ANALYTICAL LABORATORY

Sample Run Log – Total Dissolved Solids (TDS)

Standard Method 2540 C

Analyst: _____

Lab No.	Vol. Filtered (mL)	Dish No	Initial Wt Date	Initial Wt (gm)	Final Wt (1) Date	Final Wt (1) (gm)	Final Wt (2) Date	Final Wt. (2) (gm)	Net Wt (gm)	TDS (mg/L)	Average & %RPD
DI Blank											
QC											

Particulate Phosphorus

EPA Method 365.1

1.0 SCOPE AND APPLICATION

1.1 This method is applicable to the determination of particulate phosphorus in surface and saline waters.

1.2 The applicable range is 0.05 to 1.0 mg P/L.

2.0 SUMMARY OF METHOD

2.1 Samples for particulate phosphorus are collected by filtering known volumes of water samples through the filters in the field. The filters are folded, placed in aluminum foil pouches, and kept frozen until the analysis time.

2.2 Filters are combusted at 550^{o}C for 1.5 hours and treated with 1 N hydrochloric acid for 24 hrs.

2.3 The supernatant is analyzed for orthophosphate using Lachat Method *12-115-01-1-F*, where ammonium molybdate and antimony potassium tartrate react 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.

3.0 INTERFERENCES

3.1 High iron concentrations (Fe^{3+} greater than 50 mg/L) can cause precipitation of, and subsequent loss, of phosphorus.

3.2 Silica forms a pale blue complex, which also absorbs at 880nm. This interference is generally insignificant as a silicate concentration of approximately 1 mg/L SiO_2/L would be required to produce a 0.3 µg P/L positive error in orthophosphate.

3.3 Arsenate is determined similarly to phosphorus and should be considered when present in concentrations higher than phosphorus.

3.4 Method interferences may be caused by contaminants in the reagent water, reagents, glassware, and other processing apparatus that bias analyte response.

4.0 HEALTH AND SAFETY

4.1 Good laboratory practices should be followed during reagent preparation and instrument operation.

4.2 The use of a fume hood, protective eyewear and clothing and proper gloves are required when handling acids.

5.0 EQUIPMENT AND SUPPLIES

5.1 Equipment

5.1.1 Flow injection analysis equipment (Lachat 8000 series, QuikChem FIA +), consisted of the following modules, designated to deliver and react sample and reagents in the required order and rations:

- Sampler

5.1.1.2 Multi-channel proportioning pump

5.1.1.3 Reaction unit or manifold

5.1.1.4 Colorimetric detector with a 10 mm, 800 μL glass flow cell and an 880 nm interference filter

5.1.1.5 Data system

5.1.2 Isotemp Muffle Furnace (Fisher Scientific cat. no. 10-505-10)

5.1.3 Analytical Balance

5.2 Supplies

5.2.1 Glass calibration vials (Lachat part no. 21304)

5.2.2 Test tubes, 13 x 100 mm (Fisher Scientific cat. no. 14-961-27)

5.2.3 Volumetric flasks, Class A

5.2.4 Volumetric pipettes, Class A

5.2.5 Centrifuge tubes, 50 mL, with caps (Fisher Scientific cat. no. 14-432-22)

5.2.6 Test tubes, 16 x 125 mm (Fisher Scientific cat. no. 14-961-30)

5.2.7 Sera filters (TeckniServe cat. no. 510-4055-P01)

5.2.8 Aluminum weighing pans (Fisher Scientific cat. no. 08-732)

5.2.9 Glass Microfiber Filters, Whatman GF/F; 47 mm, 0.7 μm pore size (Fisher Scientific cat. no. 1825-047)

6.0 REAGENTS AND STANDARDS

6.1 Reagents

6.1.1 Reagent Water – Use deionized (18 megohm) water when preparing all reagents and standards. Degas deionized water and all reagents, except standards, to remove dissolved gases.

6.1.2 Stock Ammonium Molybdate Solution – Dissolve 40.0 g ammonium molybdate tetrahydrate ($(NH_4)_6Mo_7O_{24}.4H_2O$) in about 800 mL deionized water in a 1 L volumetric flask. Dilute to the mark and stir for at least 4 hours. Store in a dark plastic bottle and refrigerate.

6.1.3 Stock Antimony Potassium Tartrate Solution – Dissolve 3.22 g antimony potassium tartrate trihydrate ($K(SbO)C_2H_4O_6.3H_2O$), or 3.0g antimony potassium tartrate hemihydrates, ($K(SbO)C_2H_4O_6.1/2H_2O$), in about 600 mL DI water in a 1L volumetric flask. Dilute to mark and mix. Store in a dark bottle and refrigerate,

6.1.4 Molybdate Color Reagent – Add 213 mL stock ammonium molybdate and 72.0 mL stock antimony potassium tartrate to about 500 mL of deionized water in a 1 L volumetric flask.. Dilute to the mark with deionized water and invert to mix. Degas with helium.

6.1.5 Ascorbic Acid Reducing Solution – Dissolve 60.0 g ascorbic acid in about 800 mL deionized water in a 1 L volumetric flask. Mix on a magnetic stirrer and dilute to the mark with deionized water. Prepare fresh weekly.

6.1.6 1.0 M Hydrochloric Acid (Carrier/Diluent for Standards) – Add 83.0 mL of concentrated hydrochloric acid (37%, ACS Reagent Grade, d=1.200) to about 800 mL of deionized water in a 1L volumetric flask. Dilute to mark with deionized water after cooling to room temperature. Mix well.

6.1.7 Sodium Hydroxide - EDTA Rinse Solution – Dissolve 65.0 g sodium hydroxide and 6 g tetrasodium ethylenediamine tetraaceticacid (Na_4EDTA) in about 800 deionized water in a 1 L volumetric flask. Mix on a magnetic stirrer until it is completely dissolved. Dilute to the mark with deionized water and mix.

6.2 Standards

6.2.1 Stock Standard 100 mg P/L in 1.0 M Hydrochloric Acid – Dissolve 0.4394 g potassium phosphate monobasic (KH_2PO_4) that has been dried for two hours at 110°C, in about 800 mL of 1 M hydrochloric acid (6.1.6) in a 1 L volumetric flask. Dilute to the mark with 1.0M hydrochloric acid and mix.

6.2.2 Intermediate Standard Solution (1 mg P/L) – Add 10 mL of stock standard (6.2.1) to 800 mL of 1 M hydrochloric acid (6.1.6) in a 1L volumetric flask and dilute to 1000 mL mark and mix

6.2.3 Working standards – Prepare the standards according to the following chart; dilute each with1.0 M hydrochloric acid (6.1.6) and mix.

6.2.4 Spiking solution – Use stock standard, 100 mg P/L (6.2.1) to spike 10 ml of blank (1M HCl) with 50 uL of this solution (Blank Spike)

Concentration mg N/L	**Working Standard, mL**	**Final Volume, mL**
1.0	Use 6.2.2 (1 mg P/L)	50
0.5	25	50
0.4	40	100
0.3	15	50
0.2	10	50
0.1	5	50
0.05	2.5	50
0.00	0	50

7.0 COLLECTION, PRESERVATION, AND STORAGE

Samples are collected by the field personnel on Whatman GF/F filters (47 mm; 0.7 µm pore size) by filtering known volumes of water samples through the filters. The filters are folded, placed in aluminum foil pouches, and kept frozen until analysis.

8.0 QUALITY CONTROL

8.1 A demonstration of capability study is performed by each analyst performing the test by analyzing four replicates of an external quality control sample during one or two analytical runs.

8.2 A method detection limit (MDL) study is performed once a year by analyzing seven replicates of the PP pads provided by the client, over three consecutive analytical runs. MDL is calculated as follows:

MDL = (t) x (S) where, t = Student's t value for a 99% confidence level and a standard deviation estimate with n-1 degrees of freedom [t = 3.14 for seven replicates] and S = standard deviation of the replicate analyses.
Each new analyst also performs an MDL study or when any changes in the analytical procedure or instrument are made.

8.3 In the analytical run, every tenth sample is duplicated followed a blank. The accepted value for the relative percent difference (RPD) is ± 10%.

8.4 Blank filters are processed and analyzed when provided by the field personnel.

8.5 One mid-range standard (0.40 mg P/L) is analyzed for every 10 samples.

8.6 An external quality control sample is analyzed at the beginning and at the end of each analytical run.

8.7 Acidified deionized water blank (1M HCl) is analyzed at the beginning of each analytical run and after every 10^{th} samples.

9.0 PROCEDURE

9.1 Sample Preparation

9.1.1 Place filters (samples and blanks, if provided) in labeled aluminum weighing pans and combust in a muffle furnace at 550^{o}C for 1½ hours.

.

9.1.2 Cool to ambient temperature, then transfer the combusted filters to labeled 50 mL screw cap centrifuge tubes.

9.1.3 Add 10 mL 1M hydrochloric acid to each tube.

9.1.4 Cap tubes and let stand for a minimum of 24 hours. Shake tubes several times during the 24 hour period or use the shaker for continuous vibration.

9.1.5 Pour samples into 16 x 125 mm tubes and filter using Sera filters.

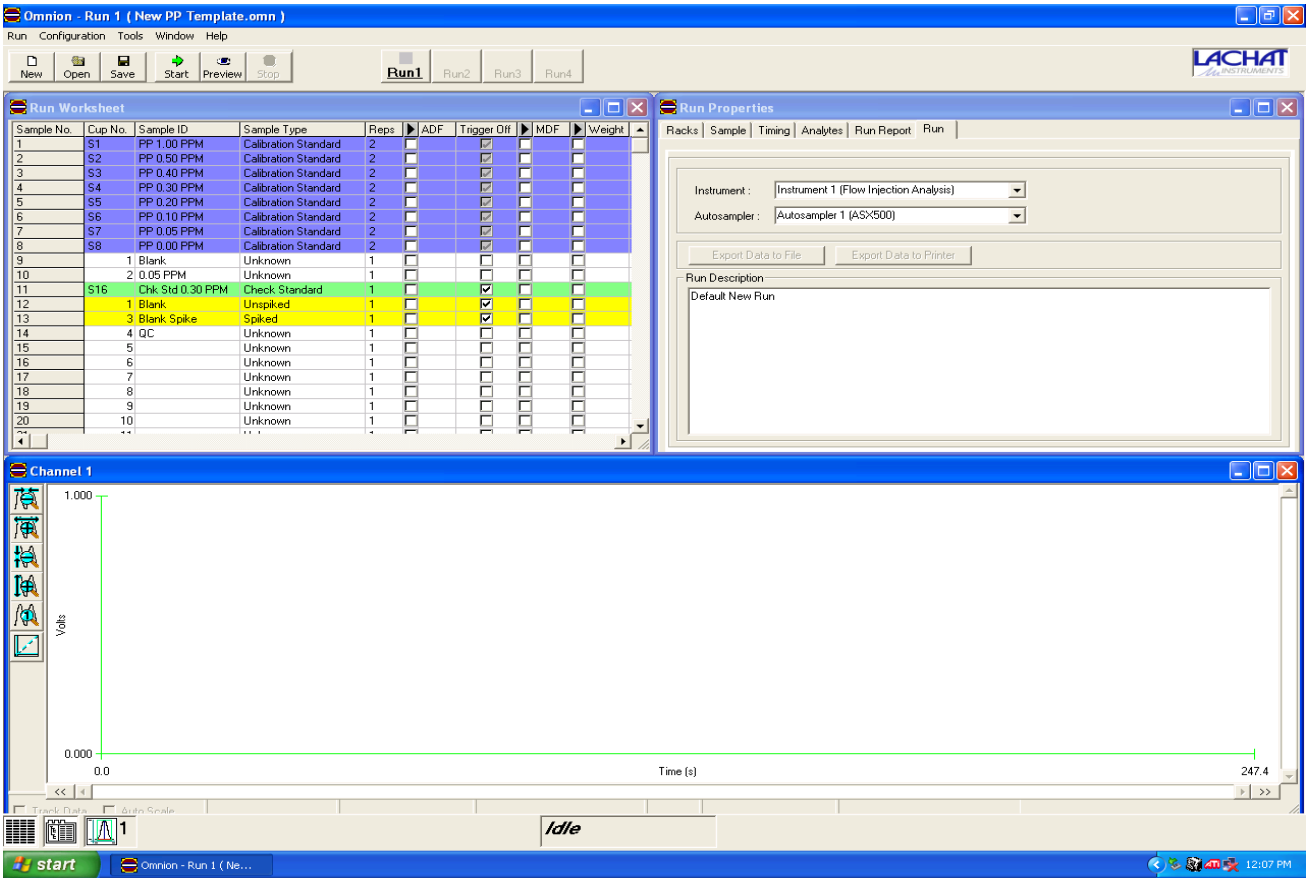
9.1.6 Transfer the filtrate to auto sampler tubes with transfer pipettes.

9.2 Instrument set-up and sample analysis

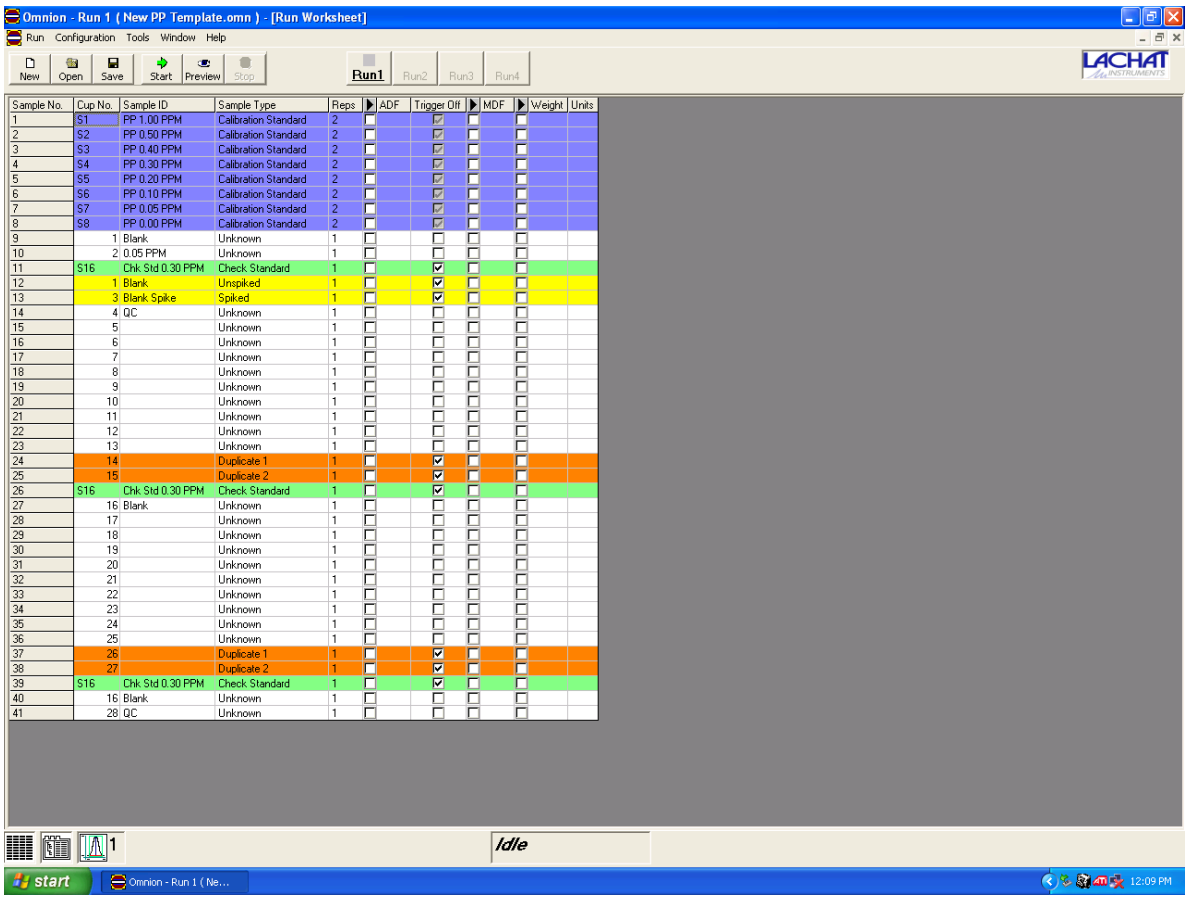
9.2.1 Set up manifold as in the attached diagram.

9.2.2 Turn on the Lachat instrument, computer, monitor, and printer.

9.2.3 Double click on Omnion and then double click on **"LL PP"** to open the template, which consists of three windows.



9.2.4 Maximize the **"Run Worksheet"** window at the top left hand corner of the screen by clicking on the middle square on that screen to open the worksheet template. Enter sample identifications in the sample identification column, making sure that all duplicates are entered in the appropriate locations of the worksheet. Identifications of the quality control samples analyzed at the beginning of the run and the locations of the duplicates and the spiked have been saved on the template. Press the **"Enter"** key after each entry in order to save all entries.

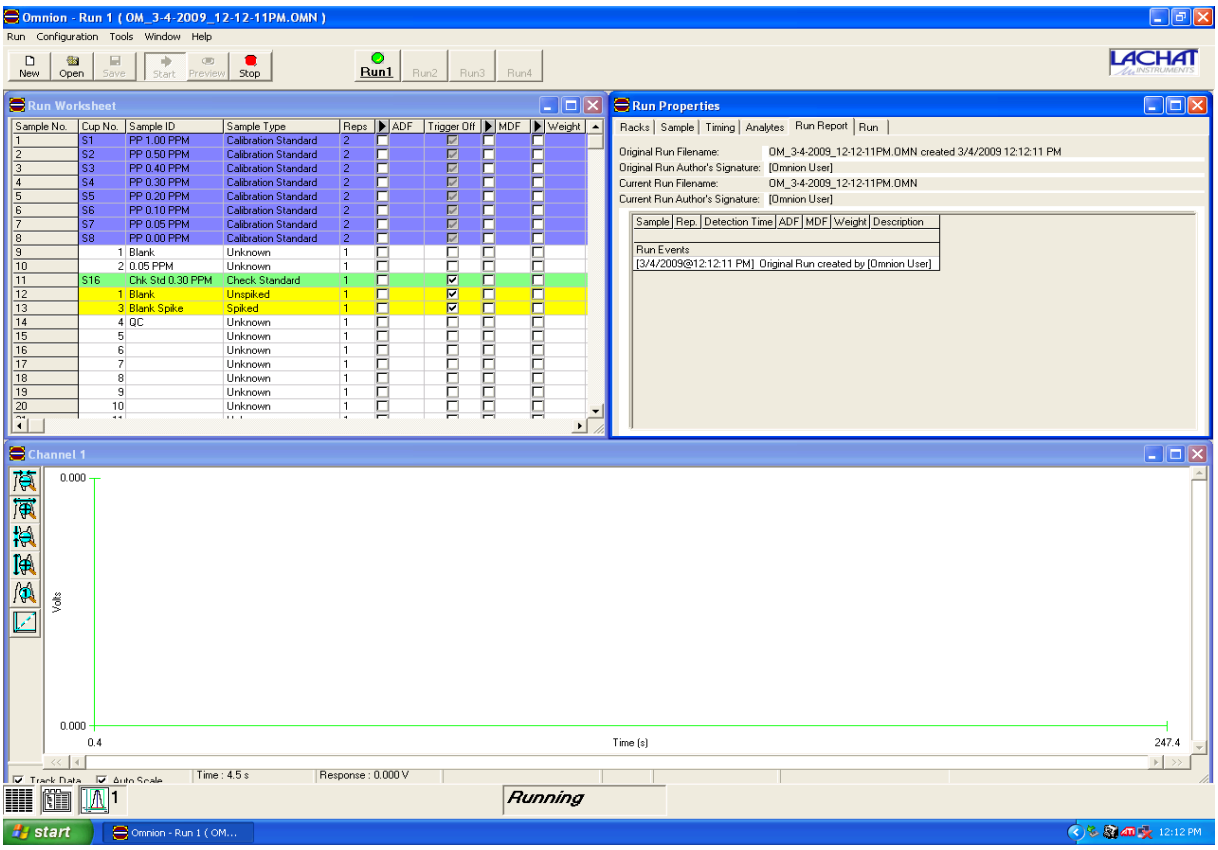


9.2.5 Print a copy of this worksheet by first double clicking on **"Run"** icon and then selecting **"Export Worksheet Data"**.

9.2.6 Click on **"Window"** tab and then, click on **"Tile"** to return to the screen with three windows.

9.2.7 Place standards in standard vials, then in the standard rack in the order of decreasing concentration from positions 1 to 8. Place the check standard vial in position 16 (S16) of the standard rack. Pour samples and quality control samples into 13 x 100 mm test tubes and place them in the sample racks according to the worksheet prepared in 9.2.4.

9.2.8 Pump deionized water through all reagent lines for 5 – 10 minutes and check for leaks and smooth flow. Switch to reagents and continue pumping for about 10 minutes. Click on **"Preview"** tab to monitor the baseline.



9.2.9 Once a stable baseline is achieved, click on **"Stop"** tab to stop monitoring the baseline. Click on **"Start"** tab to begin the analysis.

9.2.10 If the calibration passes, instrument will continue to analyze the samples. If failed, take appropriate corrective actions and recalibrate before proceeding to analyze samples.

9.2.11 Samples with concentration exceeding the calibrated range will be manually diluted by 1M HCl and reanalyzed.

9.2.12 After the run is complete, remove the reagent lines and place them in DI water and rinse for about 15 minutes. If necessary, rinse the system with the NaOH – EDTA rinse solution (6.1.7) for not more than 5 minutes followed by DI water of 10 – 15 minutes. Pump the tubes dry, release the pump tubing, and turn off the pump.

10.0 DATA ANALYSIS AND CALCULATIONS

10.1 All the calculations are performed by the Omnion 3.0 software system. The amount of color is plotted against the known concentrations and the line that best fits among the data points is the calibration curve. The concentration of unknown samples are determined automatically by plugging the amount of color (response) in the calibration curve equation. All standards are analyzed in duplicate and all data

points are used for the calibration curve. Samples with phosphorous concentrations greater than 1.00 ppm are diluted manually by 1.0 M HCl and reanalyzed.

10.2 Calculate the actual concentration of particulate phosphorus in samples (mg/L) by multiplying the concentration of sample mg /L X10mL / volume of sample (mL) used in the filtrate. Use the template below for calculations.

Particulate Phosphorus Calculation

Date:

(concentration*10)/volume of sample= result

Sample #	conc.	volume	value		Sample #	conc.	volume	value	
			#DIV/0!	###				#DIV/0!	###

10.3 Calculate the relative percentage difference for the duplicated samples as follows:

$$\% \text{ RPD} = \frac{\text{difference between the duplicates}}{\text{average of the duplicates}} \times 100$$

10.4 The reporting level for this method is the concentration of the lowest standard, which is 0.05 ppm.

10.5 Report results for all routine samples to three decimal places and for performance evaluation (PE) samples to three significant figures.

11.0 DATA AND RECORDS MANAGEMENT

11.1 The analysis data for the calibration blank, external QC and instrument performance check solution (IPC), i.e. the mid-range check standard, is kept on file with the sample analysis data.

11.2 Normal turnaround time for samples submitted to this lab is 2 to 10 days from the receipt. Results are reported in writing on the sample analysis request form.

11.3 The Division shall retain all pertinent laboratory records for each sample for a period of five years. At the conclusion of this period, clients shall be given the option to take custody of their sample records. The Division shall not be responsible for retrieving, copying, or transferring laboratory records exceeding the five years custody period.

12.0 WASTE MANAGEMENT

12.1 It is laboratory's responsibility to comply with all state, federal and local regulations governing waste management, particularly the hazardous waste identification rules and disposal restrictions, and to protect the air, water and land by minimizing and controlling all releases from fume and bench operation.

12.2 Samples and standards are poured down the drain and flushed with running water.

13.0 REFERENCES

13.1 United States Environmental Protection Agency, *Methods for Chemical Analysis of Water and Waste, EPA/600/R-93/100, Method 365.4, May 1993.*

13.2 Lachat Instruments, *Determination of Total Phosphate in Ashed Soil Samples by Flow Injection Analysis, Method 12-115-01-1-F, September 2003.*

13.3 Chesapeake Biological Laboratories, *Particulate Phosphorus Method, February 2004.*

13.4 EPA Method 365.1, *Methods for the Determination of Inorganic Substances in Environmental Samples*, August 1993.

13.5 American Public Health Association, *Standard Methods for the Examination of Water and Wastewater,* 21st Edition, Method 4500- P E, 2005.

13.6 Lachat Instruments, *Operating Manual for the Quikchem Automated Ion Analyzer*.

13.7 Division of Environmental Chemistry, DHMH-Laboratories Administration, *Quality Assurance Plan,* Revision 14.0, October 2014

13.8 Division of Environmental Chemistry, DHMH-Laboratories Adminstration, *Quality Manual,* Revision 1.0, November 2014

APPENDIX A

Division of Environmental Chemistry
INORGANICS ANALYTICAL LABORATORY

Data Review Checklist- Particulate Phosphorus

EPA Method 365.1

Lab Numbers[1]:_____ Analyst: _____

Date Collected: _____ Date Digested: _____ Date Analyzed: _____

Procedure	Acceptance Criteria	Status*	Comments
Holding Time	28 days @ –15°C		
Samples Analyzed	Within 5 working days		
Calibration Curve	Corr. Coeff. ≥ 0.9950		
Reagent Blank	< Reporting level (0.05 ppm)		
Blank Spike	1 per batch		
	Recovery = 90–110%		
Duplicates/Replicates	Every 10th sample or 1/batch, if less than 10 samples		
	RPD ≤ 10%		
Check Standard	After every 10th sample and at the end of the run		
	Recovery = 90–110%		
Decimal Places Reported	3		
Measured Values	Within calibration range (0.05– 1.00 ppm)		
Diluted Samples	Correct final calculations		
External QC[2]	Beginning and end of each run		
	Within acceptance range		
Changes/Notes	Clearly stated		

* Check (✓) if criteria are met. [1]Include beginning and ending numbers, account for gaps by bracketing

_____ _____
Analyst's Signature & Date Reviewer's Signature & Date

_____
Supervisor's Signature & Date

Reagents	ID
1M HCl	_____
Ascorbic Acid	_____
Color Reagent	_____

External QC

Identification = _____
True Value = _____ ppm
Range = _____ ppm

DETERMINATION OF TOTAL DISSOLVED NITROGEN IN ALKALINE PERSULFATE DIGESTS BY AUTOMATED COLORIMETRY

EPA Method 353.2

1.0 SCOPE AND APPLICATION

1.1 This method is applicable to seawater, brackish water, and non-saline water.

1.2 The applicable range is 0.1 to 5.0 mg N/L.

2.0 SUMMARY OF METHOD

The nitrate is quantitatively reduced to nitrite by passing the sample through a copperized cadmium column. The nitrite (reduced nitrate plus original nitrite) is then determined by diazotizing with sulfanilamide under acidic conditions followed by coupling with N-(1-naphthyl) ethylene diamine dihydrochloride (Marshall's reagent) to form a reddish-purple azo dye which is measured colorimetrically at 520 nm. Per manufacture's recommendation, water samples are digested for one hour with alkaline persulfate to oxidize all the nitrogen compounds present in the sample to nitrate (NO_3^-). A single multi-analyte standard is prepared for nitrate, nitrite and phosphorus as they are analyzed concurrently on the same instrument.

3.0 INTERFERENCES

3.1 Total carbon concentrations in excess of 20 mg C/L or Chemical Oxygen Demand (COD) concentrations in excess of 50 mg/L will result in complete oxidation of nitrogen compounds.

3.2 The presence of sulfide and chloride produced by seawater oxidation would reduce the column life as compared to non-saline samples.

3.3 Turbidity and colored samples that absorb at 520 nm will interfere in the colorimetric determination. Turbidity of the samples can be removed by filtration prior to analysis.

4.0 HEALTH AND SAFETY

4.1 Good laboratory practices should be followed during reagent preparation and instrument operation. The use of a fume hood, protective eyewear, lab coat and proper gloves are required when preparing reagents.

4.2 Sodium hydroxide, hydrochloric acid, and phosphoric acid used in this determination have the potential to be highly toxic or hazardous. Consult Material Safety Data Sheets (MSDS) for detailed explanations.

4.3 Each employee is issued a *Laboratory Safety Manual* and is responsible for adhering to the recommendations contained therein.

5.0 EQUIPMENT AND SUPPLIES

5.1 Equipment

5.1.1 Lachat QuickChem 8500 Automated Ion Analyzer, which includes the following:

5.1.1.1 Automatic sampler

5.1.1.2 Multi-channel proportioning pump

5.1.1.3 Injection module with a 12.5 cm x 0.5 mm ID sample loop

5.1.1.4 Manifold

5.1.1.5 Colorimetric detector

5.1.1.5.1 Flow cell, 10 mm path length

5.1.1.5.2 Interference filter, 520 nm

5.1.1.6 Computer with Omnion 3.0 Data system and printer

5.1.2 Analytical balance capable of accurately weighing to the nearest 0.0001 g

5.1.3 Top loading balance for weighing chemicals for reagents

5.2 Supplies

5.2.1 Class A volumetric flasks, 50 – 1,000 mL

5.2.2 Class A volumetric pipettes, 1– 10 mL

5.2.3 Automatic pipetters, 100 µL- 10 mL

5.2.4 Digestion tubes – 40 mL vials (Fisher 14-959-37C or equivalent) with autoclavable caps lined with Teflon liners (Kimble Caps Cat # 03-340-77E)

5.2.5 Beakers, disposable, polypropylene, 50 mL(Fisher 01-291-10)

5.2.6 Test tubes, glass, 13 x 100 mm and 16 X 125 mm

5.2.7 Reagent storage bottles, plastic or glass

5.2.8 Cadmium Reduction Column (Lachat Part No. 50237A)

5.2.9 Ultra High Purity Helium gas for degassing

6.0 REAGENTS AND STANDARDS

6.1 Reagents

Use deionized water (DI) for preparing all solutions. Prevent the bubble formation by degassing deionized water and all reagents with helium for two minutes.

6.1.1 15 N Sodium Hydroxide – Gradually add 150 g NaOH in a beaker of about 200 mL DI water. Ensure dissolution. Mix well, let the solution reach to room temperature, and store in a plastic container**.**

6.1.2 Ammonium Chloride Buffer, pH 8.5 – While working In a fume hood, dissolve 85.0 g ammonium chloride (NH_4Cl) and 1.0g disodium ethylenediamine tetraacetic acid dihydrate ($Na_2EDTA.2H_2O$) in about 800 mL DI water, in a 1L volumetric flask. Mix well and dilute to the mark. Adjust the pH to 8.5 with 15 N sodium hydroxide solution and then filter the reagent.

6.1.3 Sulfanilamide Color Reagent – Add about 600 mL of DI water into a 1 L volumetric flask. Then add 100 mL 85% phosphoric acid (H_3PO_4), 40.0 g sulfanilamide, and 1.0 g N- (1-naphthyl) ethylenediamine dihydrochloride (NED). Stir for about 30 minutes until dissolved. Dilute to the mark, filter and store in a dark bottle. This solution is stable for one month.

6.1.4 Digestion Solution (1Liter) – Add about 600 mL DI water into a 1 L volumetric flask. Then, add 20.1 g potassium persulfate ($K_2S_2O_8$), and 3 g sodium hydroxide (NaOH). Dilute to mark. Prepare fresh for the same day analysis per manufacturer's recommendation.

6.1.5 Borate Buffer, 1.0 M, pH 7.5 - dissolve 61.8 g boric acid and 8.0 g sodium hydroxide in about 800 mL DI water in a 1 L volumetric flask. Mix on a magnetic stirrer for about four hours until it is completely dissolved. Dilute to the mark with DI water and mix.

6.2 Standards

6.2.1 Stock Nitrate (1000 mg N/L) Standard Solution - Dissolve 0.722 g of potassium nitrate (dried in the oven for two hours) in about 60 mL of DI water in a 100 mL volumetric flask. Dilute to mark and mix. Prepare monthly.

6.2.2 Stock phosphorous (100 mg P/L) Standard Solution 0.4394 g of anhydrous potassium dihydrogen phosphate (KH2PO4), (dried in the oven for 2

hours at 110o C) in about 200 mL of DI water in a 1000 mL volumetric flask. Dilute to mark and mix. Store in a dark bottle and prepare monthly.

6.2.3 Combined Intermediate Standard Solution (1 mg P/L and 10 mg N/L) – Add 10 mL of (6.2.2) 100 mg P/L (stock standard solution for total dissolved phosphorus determination) and 10 mL of stock nitrate standard solution (6.2.1) to about 800 mL DI water in a 1L volumetric flask. Dilute to mark and mix. Prepare weekly.

6.2.4 Combined Working Standard Solutions (5.0, 2.0, 1.0, 0.5, 0.2, 0.1and 0.0 ppm) - Use the following table to prepare standards. Prepare per run and they are good for 48 hours.

Concentration, mg N/L	Working Standard, mL	Final Volume, mL
5.0	50	100
2.0	20	100
1.0	20	200
0.5	5	100
0.2	2	100
0.1	1	100
0.0	0	100

6.2.5 Stock Nitrite Standard Solution for Cadmium check (1000 mg N/L) - Dissolve 0.6072 g potassium nitrite in about 80 mL of DI water in a 100 mL volumetric flask. Dilute to mark and mix. Prepare monthly.

6.2.6 Nitrite working standard for Cadmium check (2.5ppmN/L)
Pipette 0.5 ml of 6.2.5 into 200 mL volumetric flask. Dilute to mark and mix

6.2.7 Nitrate working Standard for Cadmium check (2.5 mg N/L) – Pipette 0.5 mL of (6.2.1) to about 100 mL DI water in a 200ml volumetric flask. Dilute to mark and mix.

6.2.8 Spiking Solution – Pipette 50 µl of a combined solution of 10mL of 1000 mg/L N (6.2.1) and 10 mL of 100 mg/L P (6.2.2) into 10 mL of sample (sample spike)or 10 mL of DI water (blank spike).

7.0 SAMPLE COLLECTION, PRESERVATION, AND STORAGE

7.1 Samples are filtered in the field, collected in plastic bottles or cubitainers, and are preserved by cooling to 4^{o} C.

7.2 Samples are analyzed within 48 hrs after collection. If they cannot be analyzed within this time period, they may be frozen at -20^{o} C as soon as possible at the laboratory. The holding time for frozen samples is 28 days.

8.0 QUALITY CONTROL

8.1 An initial Demonstration of Performance (IDP) to establish the instrument's capability is required prior to its use in analyzing client's samples. The linearity of calibration range (LCR) and the ability to quantify correctly Quality Control Samples are used to assess performance.

8.2 A mid-range check standard and a calibration blank is analyzed immediately following daily calibration, after every tenth sample (or more frequently, if required) and at the end of the sample run. The acceptable concentrations for the check standard must be within ± 10% of the actual concentration of the check standard. If a check standard fails, the samples between the last satisfactory check standard and the unsatisfactory standard are re-analyzed. Blank concentration must be less than the reporting level of 0.1 ppm. Blanks that do not meet this criterion are reanalyzed.

8.3 Every tenth sample is duplicated (analyzed from two different tubes) and spiked. The accepted values for the relative percent difference (RPD) must fall within ± 10 % and for spike recovery between 90 - 110%. If these do not fall within the accepted ranges, the corresponding analyses are repeated.

8.4 A QC sample with a known concentration and a range is analyzed at the beginning and at the end of each run. QC samples that do not fall within the accepted range are repeated.

8.5 Samples with a concentration exceeding the calibrated range are diluted manually and reanalyzed.

8.6 Data acceptance criteria are listed on the data review checklist (page 15)

8.7 The laboratory annually participates in USGS, Chesapeake Bay Laboratory (CBL), Water Supply (WS) and Water Pollution (WP) proficiency studies.

8.8 A method detection limit (MDL) study is performed once a year by analyzing seven replicates of the 0.1 ppm standard spread over three consecutive analytical runs. MDL is calculated as follows:

MDL = (t) x (S) where, t = Student's t value for a 99% confidence level and a standard deviation estimate with n-1 degrees of freedom [t = 3.14 for seven replicates] and S = standard deviation of the replicate analyses.
Each new analyst also performs an MDL study or when any changes in the analytical procedure or instrument are made.

8.9 A demonstration of capability study is performed by each analyst performing the test by analyzing four replicates of an external quality control sample during one or two analytical runs.

8.10 The efficiency of the cadmium column before and after sample run is calculated by running 2.5 ppm NO_2-N (6.2.6) and 2.5 ppm NO_3-N (6.2.7) standards and using the formula (NO_3-N/NO_2-N) x 100. The accepted range for the cadmium column efficiency is 90-110%. If the efficiency is out of this range, new standards are prepared and efficiency is re-evaluated. If the efficiency is still out of range then the column is replaced.

9.0 PROCEDURE

9.1 Sample preparation

9.1.1 Make a list of samples to be analyzed and pour aliquots of samples into labeled 16 mm x 125 mm test tubes.

9.1.2 Pipette 10 mL of each standard or sample into digestion tubes.

9.1.3 Pipette 10 mL of a mid-range (0.1 mg P/L and 1.0 mg N/L) standard, a blank, a blank spike, and an external quality control sample into digestion tubes. With each tray prepare a duplicate and a spike of every 10^{th} sample.

9.1.4 Pipette 10 mL of the nitrate and nitrite standards for cadmium column check (6.2.6 and 6.2.7) into digestion tubes.

9.1.5 Add 5 mL of digestion solution to each tube, screw the caps on tightly and mix each. Digest the standards, samples, and all the quality control samples in the autoclave for 60 min. at 121 ^{o}C (250 ^{o}F) @ 17 psi.

For Autoclave Operation please see the manual

9.1.6 After one hour, turn off the autoclave and let the digests cool for 5 minutes. Remove the tubes from the autoclave and let the digests come to room temperature. Add 1 mL of borate buffer, vortex, and pour into 13 x 100 mm culture tubes analyze them as soon as possible.

9.1.7 If samples cannot be analyzed same day, do not add the borate buffer. Refrigerate the digests at 4^{o}C. Refrigerated digests will be brought up to room temperature, and subsequently 1 mL borate buffer (6.1.5) is added to each tube and mixed thoroughly by a vortex.

9.1.8 Analyze the digests using the procedure described in 9.2.

9.2 Instrument Calibration and Sample Analysis

9.2.1 Set up manifold according to the manifold diagram.

9.2.2 Pump deionized water through all reagent lines and check for leaks and a smooth flow. Switch to reagents and allow the system to stabilize and a stable baseline is achieved.

9.2.3 Enter sample information required by the data system.

9.2.4 Place standards, blanks, samples, quality controls, etc. in the auto sampler according to the run table.

9.2.5 Initiate the analytical run.

9.2.6 At the end of the run, review the calibration curve statistics and the results for the quality control samples. Acceptable values for the correlation coefficient are ≥ 0.9950. Other quality control criteria are described in 8.0.

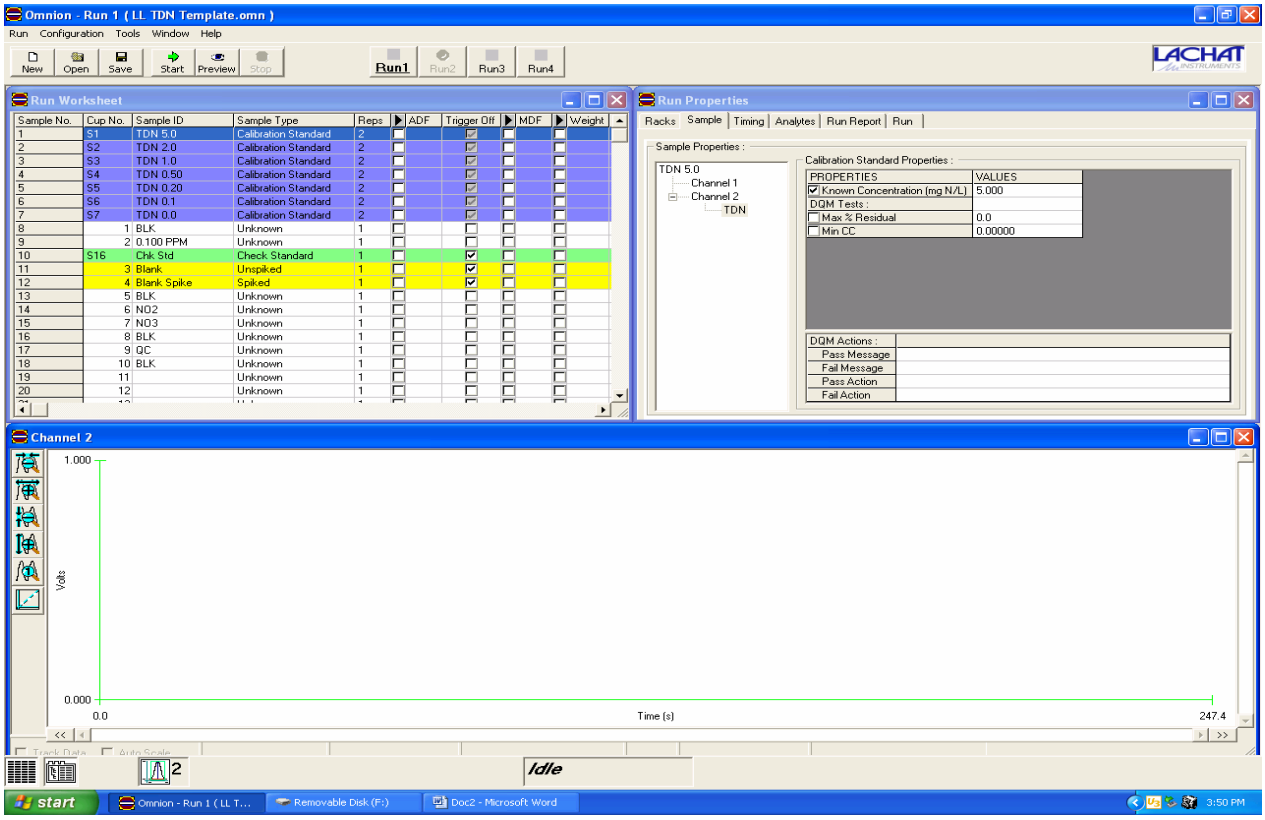
9.2.7 Get the data reviewed by a designated scientist, and then, report the results on the Analysis Request Forms.

9.3 Instrument set-up and sample analysis

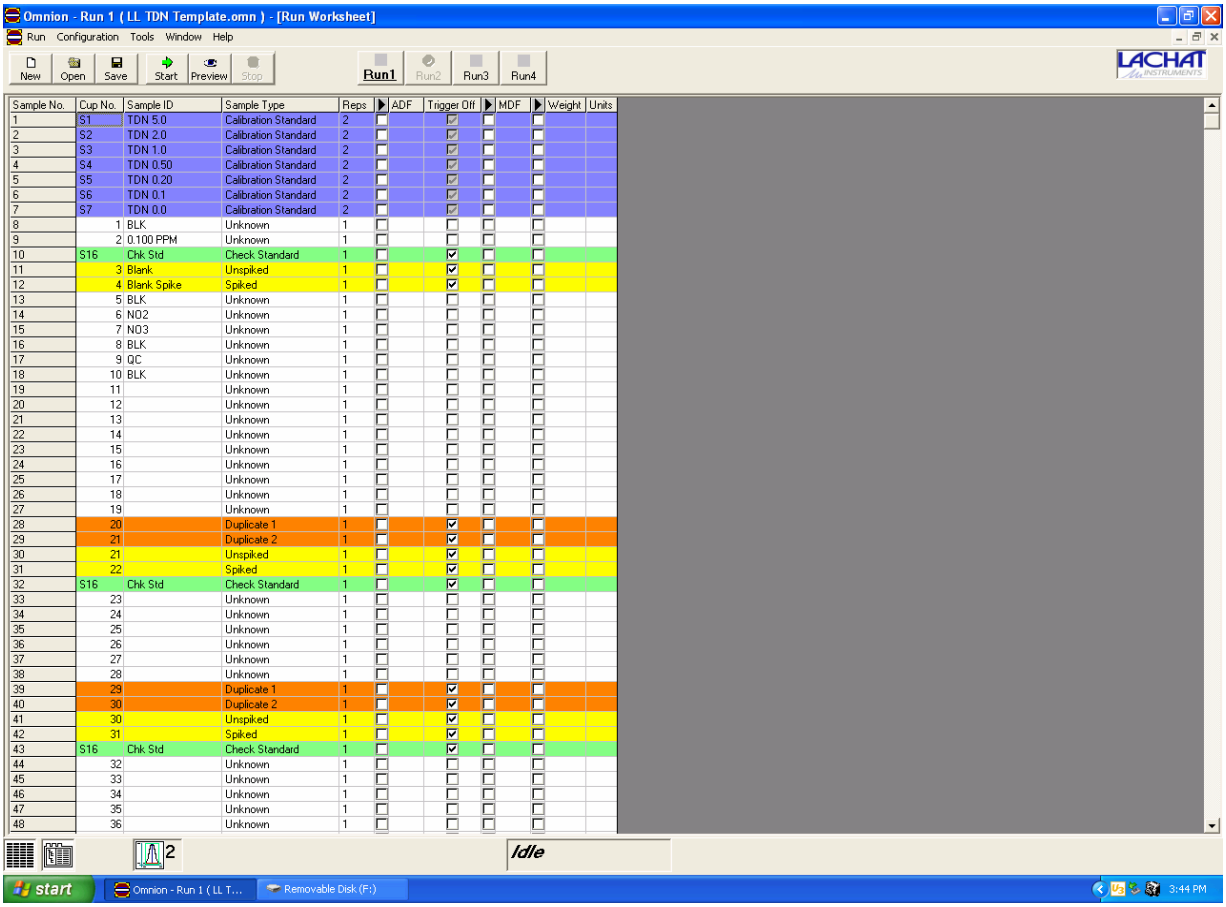
9.3.1 Set up manifold as in the diagram.

9.3.2 Turn on the Lachat instrument, computer, monitor, and printer.

9.3.3 Double click on Omnion and then on **"LL TDN"** to open the template, which consists of three windows.



9.3.4 Maximize the **"Run Worksheet"** window at the top left hand corner of the screen by clicking on the middle square on that screen to open the worksheet template. Enter sample identifications in the sample identification column, making sure that all duplicates and spikes are entered in the appropriate locations of the worksheet. Identifications of the quality control samples analyzed at the beginning of the run and the locations of the duplicates and the spiked have been saved on the template. Make sure that **"Enter"** key is pressed after each entry in order to save all entries.

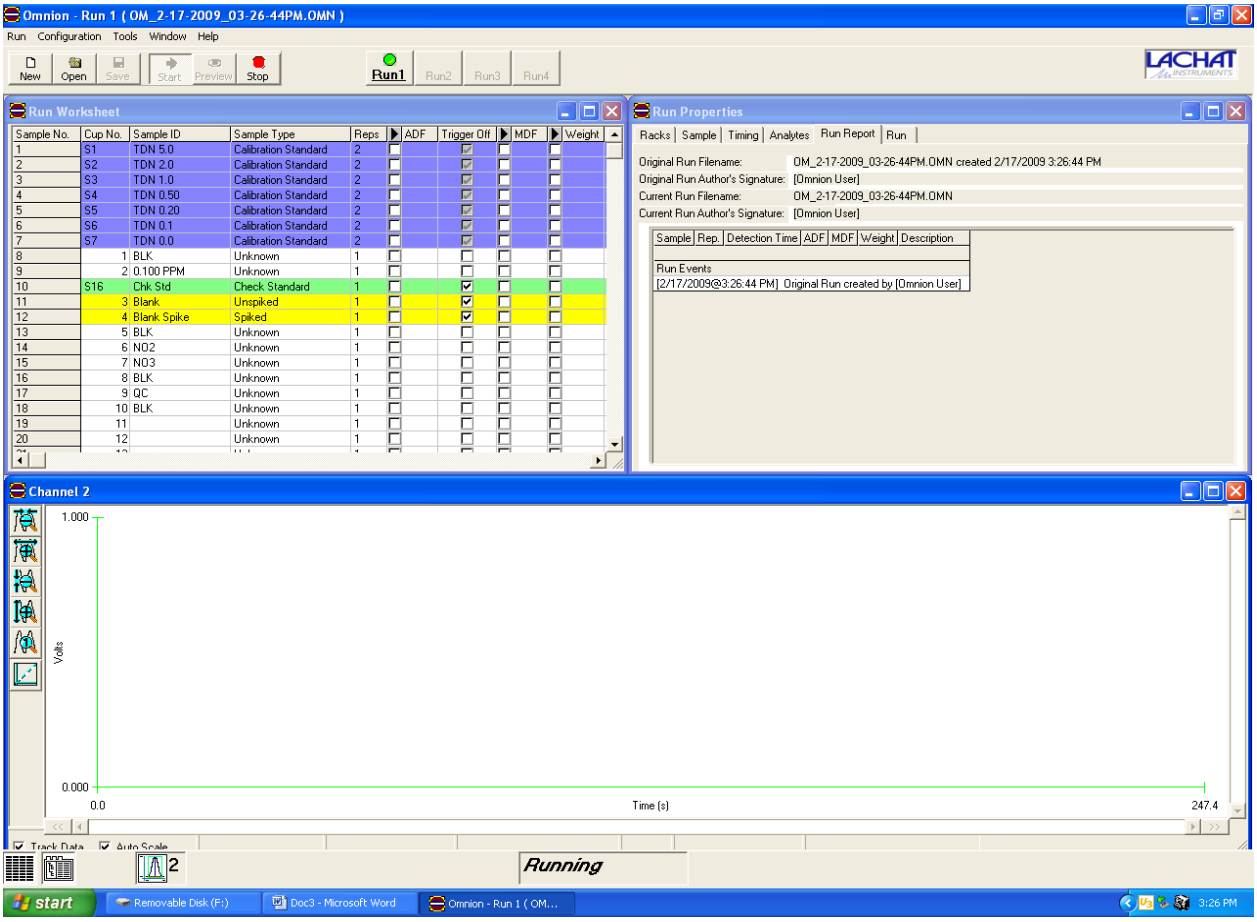


9.3.5 Print a copy of this worksheet by first double clicking on **"Run"** icon and then selecting **"Export Worksheet Data"**.

9.3.6 Click on **"Window"** tab and then, click on **"Tile"** to return to the screen with three windows.

9.3.7 Place standards in standard vials in the standard rack in order of decreasing concentration in positions 1 to 7 (STD 7 is DI water -0.00 mg/L). Place the check standard vial in position 16 (S16) of the standard rack. Pour samples and quality control samples into 13 x 100 mm test tubes and place them in the sample racks according to the worksheet.

9.3.8 Pump deionized water through all reagent lines for 10 – 15 minutes and check for leaks and smooth flow. Switch to reagents and continue pumping for about 10 minutes. Click on **"Preview"** tab to monitor the baseline.



9.3.9 Once a stable baseline is achieved, click on **"Stop"** tab to stop monitoring the baseline. Click on **"Start"** tab to begin the analysis.

9.3.10 If the calibration passes, curve fit of calibration solutions/response based on regression, the instrument will continue to analyze the samples. If it fails, take appropriate corrective actions and recalibrate before proceeding to analyze samples.

9.3.11 Manual dilution will be performed to reanalyze samples with concentrations exceeding the calibrated range.

9.3.12 After the run is complete, remove the reagent lines and place them in DI water and rinse for about 15 minutes. If necessary, rinse the system with the NaOH – EDTA rinse solution for not more than 5 minutes, followed by DI water rinse for 10 – 15 minutes. Pump the tubes dry, release the pump tubing, and turn off the pump.

10.0 DATA ANALYSIS AND CALCULATIONS

10.1 All the calculations are performed by the Omnion 3.0 software system. The amount of color is plotted against the known concentrations and the line that best

fits among the data points is the calibration curve. The concentration of unknown samples is determined automatically by plugging the amount of color (response) in the calibration curve equation. All standards are analyzed in duplicate and all data points are used for the calibration curve. Samples with total dissolved nitrogen concentrations greater than 5.00 ppm are diluted manually by 1.0 M HCl and reanalyzed.

10.2 Calculate the percentage spike recovery of the laboratory fortified samples as follows:

$$\% \text{ SR} = \frac{(\text{spiked sample conc.} - \text{sample conc.}),\ \text{ppm}}{\text{amount of spike added to sample, ppm}} \times 100$$

10.3 Calculate the relative percentage difference for the duplicated samples as follows:

$$\% \text{ RPD} = \frac{\text{difference between the duplicates}}{\text{average of the duplicates}} \times 100$$

10.4 The reporting level for this method is the concentration of the lowest standard, which is 0.1 ppm.

10.5 Report results for all routine samples to three decimal places and for performance evaluation (PE) samples to three significant figures.

11.0 DATA AND RECORDS MANAGEMENT

11.1 The analysis data for the calibration blank, external QC and instrument performance check solution (IPC), i.e. the mid-range check standard, is kept on file with the sample analysis data.

11.2 Normal turnaround time for samples submitted to this lab is 2 to 10 days from the receipt. Results are reported in writing on the sample analysis request form.

11.3 The Division shall retain all pertinent laboratory records for each sample for a period of five years. At the conclusion of this period, clients shall be given the option to take custody of their sample records. The Division shall not be responsible for retrieving, copying, or transferring laboratory records exceeding the five years custody period.

12.0 WASTE MANAGEMENT

12.1 It is laboratory's responsibility to comply with all state, federal and local regulations governing waste management, particularly the hazardous waste identification rules and disposal restrictions, and to protect the air, water and land by minimizing and controlling all releases from fume and bench operation.

12.2 Samples and standards are poured down the drain and flushed with running water.

13.0 REFERENCES

13.1 EPA Method 353.2, *Methods for the Determination of Inorganic Substances in Environmental Samples*, August 1993.

13.2 American Public Health Association, *Standard Methods for the Examination of Water and Wastewater,* 21st Edition, Method 4500- P E, 2005.

13.3 Lachat Instruments, *Methods Manual for the Quikchem Automated Ion Analyzer,* Method 10-107-04-4-A.

13.4 Lachat Instruments, *Operating Manual for the Quikchem Automated Ion Analyzer*.

13.5 Division of Environmental Chemistry, DHMH-Laboratories Administration, *Quality Assurance Plan,* Revision 14.0, October 2014

13.6 Division of Environmental Chemistry, DHMH-Laboratories Administration, *Quality Manual,* Revision 1.0, November 2014

APPENDIX A

Division of Environmental Chemistry
INORGANICS ANALYTICAL LABORATORY

Data Review Checklist Total Dissolved Nitrogen (TDN)/Alkaline Persulfate Digestion

EPA Method 353.2

Lab Numbers[1]:_____ Analyst: _____

Date Collected: _____ Date Digested: _____ Date Analyzed: _____

Procedure	Acceptance Criteria	Status*	Comments
Holding Time	48 hours @ 4°C; 28 days @ –20°C		
Calibration Curve	Corr. Coefficient. ≥ 0.9950		
Reagent Blank	< Reporting level (0.100 ppm)		
Blank Spike	1 per batch		
	Recovery = 90–110%		
Matrix Spike	Every 10th sample or 1/batch, if less than 10		
	Recovery = 90–110%		
External QC	Beginning and end of each run		
	Within acceptance range		
Check Standard	After every 10th sample and at the end of the run		
	Recovery = 90–110%		
Duplicates/Replicates	Every 10th sample or 1/batch, if less than 10 samples		
	RPD ≤ 10%		
Cadmium Column Check	90-110%		
Decimal Places Reported	3		
Measured Values	Within calibration range (0.100–5.00 ppm)		
Diluted Samples	Correct final calculations		
Changes/Notes	Clearly stated		

[1]Include beginning and ending numbers, account for gaps by bracketing.

_____ _____
Analyst's Signature & Date Reviewer's Signature & Date

_____
Supervisor's Signature and Date

Reagents	ID	Reagents	ID	External QC	
Ammonia Buffer	_____	Oxidizing Reagent	_____	Identification =	_____
Color Reagent	_____	Borate Buffer	_____	True Value =	_____ ppm
				Range =	_____ ppm

APPENDIX B

Division of Environmental Chemistry
INORGANICS ANALYTICAL LABORATORY
Data Review Checklist LL Total Dissolved Phosphorus TDP/ LL Total Dissolved Nitrogen TDN
Chesapeake Bay Program Method

Lab Numbers: [1] _____ Analyst: _____

_____ Date Digested: _____

Dates Collected: _____ Date Analyzed _____

Procedure	Acceptance Criteria	Status (√)	Comments
Holding Time	48 hours @ 4°C; 28 days @ –20°C		
Calibration Curve	Corr. Coeff. ≥ 0.9950		
Reagent Blank	< Reporting Level (0.010 ppm for TDP; 0.100 ppm for TDN)		
Blank Spike	1 per batch		
	Recovery = 90–110%		
Matrix Spike	Every 10th sample or 1/batch, if less than 10 samples		
	Recovery = 90–110%		
External QC	Beginning and end of each run		
	Within acceptable range		
Check Standard	After every 10th sample and at the end of the run		
	Recovery = 90–110%		
Duplicates/Replicates	Every 10th sample or 1/batch, if less than 10 samples		
	RPD ≤ 10%		
Cadmium Column Check	90-110%		
Decimal Places Reported	3		
Measured Values	Within calibration range (0.010–0.500 ppm for TDP; 0.100–5.000 ppm for TDN)		
Diluted Samples	Correct final calculations		
Changes/Notes	Clearly stated		

[1]Include beginning and ending numbers, account for gaps by bracketing.

_____ _____
Reviewer's Signature and Date Date Reported

_____ _____
Supervisor's Signature and Date Analyst's Signature and Date

Reagents	ID	Reagents	ID
Ammonia Buffer		Sulfanilamide	
Ascorbic Acid		Color Reagent	
Borate Buffer		Molybdate	
1M HCl		Color Reagent	
Oxidizing Reagent			

External QC

Identification = _____

True Value = _____ TDN /TDP _____ ppm

Range = TDN _____ ppm

Range = TDP _____ ppm

TOTAL DISSOLVED PHOSPHORUS IN ALKALINE PERSULFATE DIGESTS

EPA Method 365.1

1.0 SCOPE AND APPLICATION

1.1 This method is applicable to seawater, brackish water, and non-saline water.

1.2 The applicable range is 0.01 to 0.5 mg P/L.

2.0 SUMMARY OF METHOD

Water samples are digested for one hour with alkaline persulfate to convert all of the phosphorus present in the sample to orthophosphate (PO_4^{3-}). Approximately 1.3 mL of this digest is injected onto the manifold, where orthophosphate reacts with ammonium molybdate and antimony potassium tartrate under acidic condition and then reduced with ascorbic acid to form a blue complex which absorbs light at 880 nm. Per manufacture's recommendation, water samples are digested for one hour with alkaline persulfate to oxidize all the phosphorus compounds present in the sample to orthophosphate (PO_4^{3-}). The absorbance is directly proportional to the concentration of phosphorus in the sample.

3.0 INTERFERENCES

3.1 Silica forms a pale blue complex which also absorbs at 880 nm. A silica concentration of 4000 ppm would produce a 1 ppm positive error in orthophosphate.

3.2 Glassware should be washed with 1:1 HCl and rinsed with deionized water in order to prevent possible contamination problems in low level phosphorus determinations.

4.0 HEALTH AND SAFETY

4.1 Good laboratory practices should be followed during reagent preparation and instrument operation.

4.2 The use of a fume hood, protective eyewear, lab coat and proper gloves must be used when preparing reagents.

4.3 Sodium hydroxide, hydrochloric acid, and sulfuric acid used in this determination have the potential to be highly toxic or hazardous. Consult MSDS for detailed explanations.

5.0 EQUIPMENT AND SUPPLIES

5.1 Equipment

5.1.1 Lachat QuickChem 8500 Automated Ion Analyzer, which includes the following:

5.1.1.1 Automatic sampler

5.1.1.2 Multi-channel proportioning pump

5.1.1.3 Injection module with a 12.5 cm x 0.5 mm ID sample loop

5.1.1.4 Manifold

5.1.1.5 Colorimetric detector

5.1.1.5.1 Flow cell, 10 mm path length

5.1.1.5.2 Interference filter, 880 nm

5.1.1.6 Computer with Omnion 3.0 Data system and printer

5.1.2 Analytical balance capable of accurately weighing to the nearest 0.0001 g

5.1.3 Top loading balance for weighing chemicals for reagents

5.2 Supplies

5.2.1 Class A volumetric flasks, 50 – 1,000 mL.

5.2.2 Class A volumetric pipettes, 1– 10 mL.

5.2.3 Automatic pipetters, 100 µL- 10 mL

5.2.4 Digestion tubes – 40 mL vials (Fisher 14-959-37C or equivalent) with autoclavable caps lined with Teflon liners (Kimble Caps Cat # 03-340-77E)

5.2.5 Beakers, disposable, polypropylene, 50 mL(Fisher 01-291-10)

5.2.6 Test tubes, glass, 13 x 100 mm and 16 X 125 mm

5.2.7 Reagent storage bottles, plastic or glass

5.2.8 Ultra High Purity Helium gas for degassing

6.0 REAGENTS AND STANDARDS

6.1 Reagents

Use deionized water (DI) for preparation of all solutions. Prevent the bubble formation by degassing deionized water and all reagents except the standards with helium for two minutes.

6.1.1 Alkaline Persulfate Oxidizing Reagent - In a 1L volumetric flask, dissolve 20.1 g potassium persulfate ($K_2S_2O_8$), and 3g sodium hydroxide (NaOH) in about 600 mL DI water. Dilute to mark and mix. Prepare fresh daily before use.

6.1.2 Hydrochloric Acid, 1.0 M – Add 83.3 mL concentrated hydrochloric acid (37%, ACS Reagent Grade, d = 1.200) to about 800 mL of DI water in a 1L volumetric flask in a fume hood. Dilute to mark and mix well.

6.1.3 Stock Ammonium Molybdate Solution – Dissolve 40.0 g ammonium molybdate tetrahydrate ($(NH_4)_6Mo_7O_{24}$, 4H2O) in about 800 mL DI water in a 1 L volumetric flask. Dilute to the mark and stir until completely dissolved; this may take about 4 hours. Store in plastic and refrigerate.

6.1.4 Stock Antimony Potassium Tartrate Solution – Dissolve 3.22 g antimony potassium tartrate trihydrate ($K(SbO)C_2H_4O_6.3H_2O$) in about 600 mL DI water in a 1L volumetric flask. Dilute to mark and mix. Store in a dark bottle and refrigerate.

6.1.5 Molybdate Color Reagent – In a hood, carefully add 70.0 mL concentrated sulfuric acid to about 500 mL water in a 1 L volumetric flask and mix well. Then, add 72.0 mL stock antimony potassium tartrate (6.1.4) and 213 mL stock ammonium molybdate (6.1.3). Dilute to the mark with DI water. Degas with helium.

6.1.6 Ascorbic Acid Reducing Solution – Dissolve 75.0 g ascorbic acid in about 800 DI water in a 1 L volumetric flask. Mix on a magnetic stirrer and dilute to the mark with DI water. Prepare fresh weekly.

6.1.7 Borate Buffer, 1.0 M, pH 7.5 - Dissolve 61.8 g boric acid and 8.0 g sodium hydroxide in about 800 mL DI water in a 1 L volumetric flask. Mix on a magnetic stirrer until it is completely dissolved. Dilute to the mark with DI water.

6.1.8 Carrier Solution - Combine 300 mL of oxidizing reagent (6.1.1), 60.0 mL 1 N hydrochloric acid (6.1.2), and 60.0 mL borate buffer (6.1.7) in a 1 L volumetric flask, dilute to volume, and stir well. Degas the solution with helium. It is recommended that the carrier is degassed within 4 hours of use and prepared same day of analysis.

6.1.9 Sodium Hydroxide – EDTA Rinse - In a 1L flask, dissolve 65.0 g sodium hydroxide (NaOH) and 6 g tetrasodium ethylenediamine tetraacetic acid (Na_4EDTA) in about 800 deionized water. Mix on a magnetic stirrer until it is completely dissolved. Dilute to the mark with deionized water.

6.2 Standards

6.2.1 Stock Standard Solution (100 mg P/L) - Dissolve 0.4394 g of anhydrous potassium dihydrogen phosphate (KH_2PO_4) which has been dried for two hours at 110^{o}C in about 800 mL deionized water. Dilute to the mark and invert to mix. Prepare monthly.

6.2.2 Combined Intermediate Standard Solution (1 mg P/L and 10 mg N/L) - Add 10 mL of stock standard (6.2.1) and 10 mL of 1000 mg N/L (stock standard solution for total dissolved nitrogen determination) to about 800 mL DI water in a 1L volumetric flask. Dilute to mark and mix. Prepare weekly.

6.2.3 Spiking Solution - Use the mixed intermediate standard (6.2.2) as the spiking solution. Spike 10 mL of blanks and samples with 50 µl of this solution for blank and sample spikes.

6.2.4 Combined Working Standard Solutions - Use the following table to prepare standards. Dilute each to100 mL and mix well. DI water is used as the last standard (0.00 ppm).

Concentration mg P/L	Working Standard, ml	Final Volume, ml
0.5	50	100
0.2	20	100
0.1	20	200
0.05	5	100
0.02	2	100
0.01	1	100
0.00	0	100

7.0 SAMPLE COLLECTION, PRESERVATION, AND STORAGE

7.1 Samples are filtered in the field, collected in plastic bottles or cubitainers and preserved by cooling to 4^{o} C.

7.2 Samples are analyzed within 48 hrs after collection. If they cannot be analyzed within this time period, they may be frozen at -20^{o} C as soon as possible at the laboratory. The holding time for frozen samples is 28 days.

8.0 QUALITY CONTROL

8.1 An initial Demonstration of Performance (IDP) to establish the instrument's capability is required prior to its use in analyzing client's samples. The linearity of calibration range (LCR) and the ability to quantify correctly Quality Control Samples are used to assess performance.

8.2 A mid-range check standard and a calibration blank are analyzed Immediately following daily calibration, after every tenth sample (or more frequently, if required) and at the end of the sample run. The acceptable concentrations for the check standard must be within ± 10% of the true value. If a check standard fails, the samples between the last satisfactory check standard and the unsatisfactory standard are re-analyzed. Blank concentration must be less than the reporting level of 0.01 ppm. Blanks that do not meet this criterion are reanalyzed.

8.3 Every tenth sample is duplicated (analyzed from two different tubes) and spiked. The accepted values for the relative percent difference (RPD) must fall within ± 10 % and for spike recovery between 90 - 110%. If these do not fall within the accepted ranges, the corresponding analyses are repeated.

8.4 A QC sample with a known concentration and a range is analyzed at the beginning and at the end of each run. QC samples that do not fall within the accepted range are repeated.

8.5 Samples with a concentration exceeding the calibrated range are diluted manually and reanalyzed.

8.6 Data acceptance criteria are listed on the data review checklist (Appendix A).

8.7 The laboratory annually participates in USGS, CBL, ERA Water Supply (WS) and Water Pollution (WP) proficiency studies.

8.8 A demonstration of capability study is performed by each analyst performing the test by analyzing four replicates of an external quality control sample during one or two analytical runs.

8.9 A method detection limit (MDL) study is performed once a year by analyzing seven replicates of the 0.01 ppm standard spread over three analytical runs. MDL is calculated as follows:

MDL = (t) x (S) where, t = Student's t value for a 99% confidence level and a standard deviation estimate with n-1 degrees of freedom [t = 3.14 for seven replicates] and S = standard deviation of the replicate analyses.
Each new analyst also performs an MDL study or when any changes in the analytical procedure or instrument are made

9.0 PROCEDURE

9.1 Sample preparation

9.1.1 Make a list of samples to be analyzed and pour aliquots of samples into labeled 16 mm x 125 mm test tubes.

9.1.2 Pipette 10 mL of each standard or sample into digestion tubes.

9.1.3 Pipette 10 mL of a mid-range standard (0.3 mg P/L and 3.0 mg N/L), a blank, a blank spike, and an external quality control sample into digestion tubes with each tray of 24 samples. Prepare a duplicate and a spike of every 10^{th} sample.

9.1.4 Pipette 10 mL of the nitrate and nitrite standards for cadmium column check (6.2.5 and 6.2.6) into digestion tubes.

9.1.5 Add 5 mL of Alkaline Persulfate Oxidizing Reagent (6.1.1) to each tube, screw the caps on tightly and mix each. Digest the standards, samples, and all the quality control samples in the autoclave for 60 min. at 121 ^{o}C (250 ^{o}F) @ 17 psi. *Please see the manual for Autoclave Operation.*

9.1.6 After one hour, turn off the autoclave and let the digests cool for 5 minutes. Remove the tubes from the autoclave and let the digests come to room temperature. Add 1 mL of borate buffer, vortex, and pour into 13 x 100 mm culture tubes analyze them as soon as possible.

9.1.7 If samples cannot be analyzed same day, do not add the borate buffer, refrigerate the digests at 4^{o}C. Refrigerated digests will be brought up to room temperature, and then 1 mL borate buffer (6.1.7) is added to each tube and mixed.

9.1.8 Analyze the digests using the procedure described in 9.2.

9.2 Instrument Calibration and Sample Analysis

9.2.8 Set up manifold according to the manifold diagram.

9.2.9 Pump deionized water through all reagent lines and check for leaks and a smooth flow. Switch to reagents and allow the system to stabilize and a stable baseline is achieved.

9.2.10 Enter sample information required by the data system.

9.2.11 Place standards, blanks, samples, quality controls, etc. in the auto sampler according to the run table.

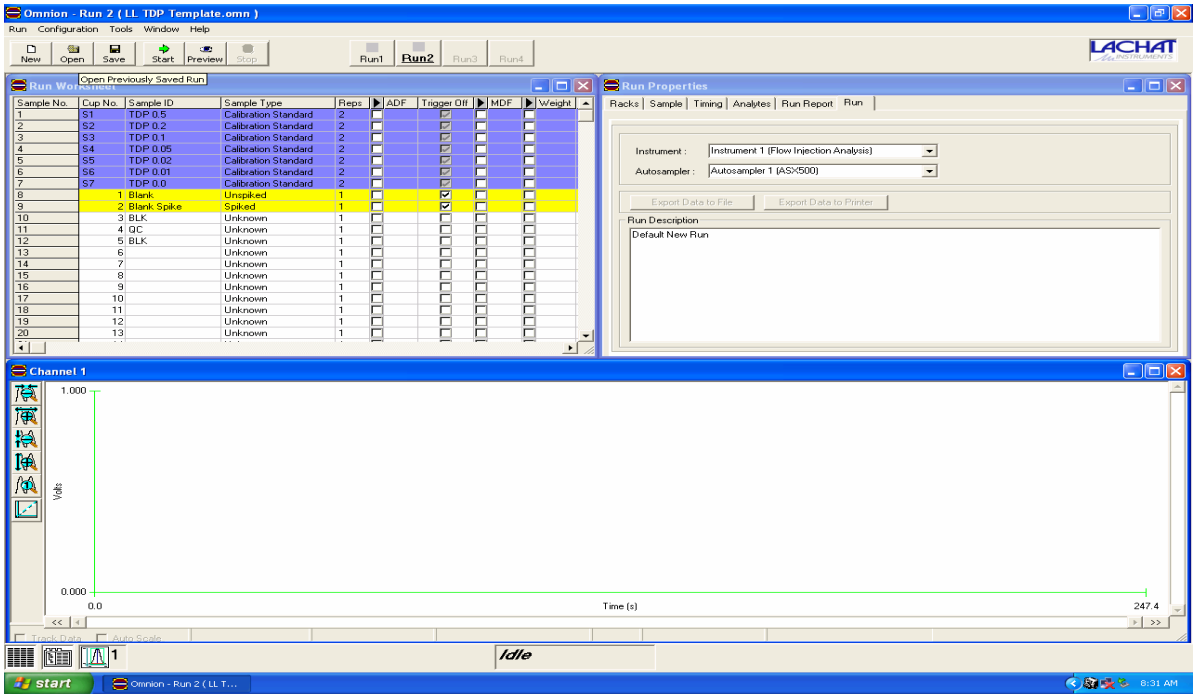
9.2.12 Click on **"Start"** tab to begin the analysis.

9.3 Instrument set-up and sample analysis

9.3.1 Set up manifold as in the diagram.

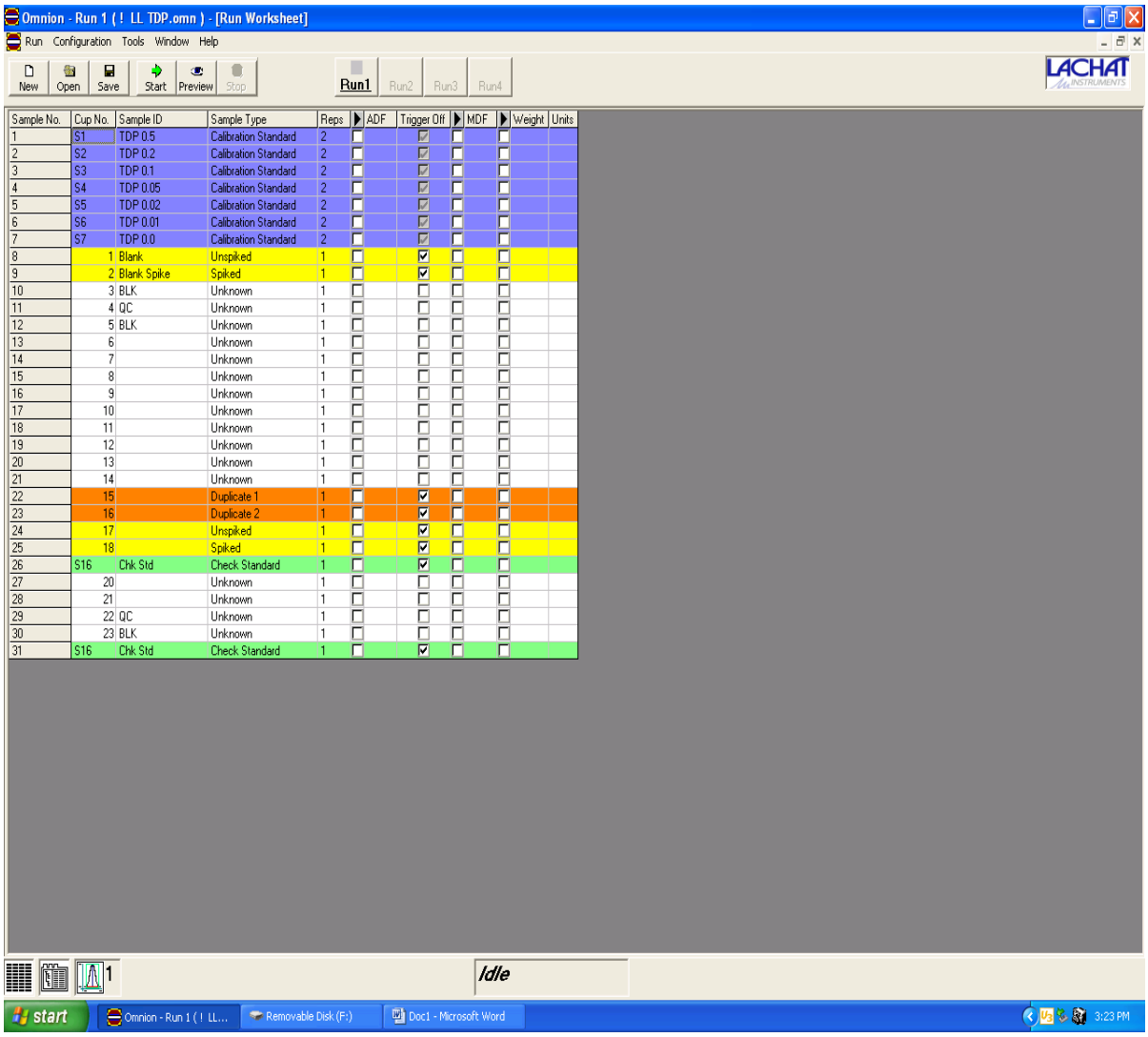
9.3.2 Turn on the Lachat instrument, computer, monitor, and printer.

9.3.3 Double click on the short-cut for **"LL TDP"** to open the template, which consists of three windows.



9.3.4 Maximize the **"Run Worksheet"** window at the top left hand corner of the screen by clicking on the middle square on that screen to open the worksheet template. Enter sample identifications in the sample ID column, making sure that all duplicates and spikes are entered in the appropriate locations of the worksheet. Identifications of the quality control samples analyzed at the beginning of the run and the locations of

the duplicates and the spiked have been saved on the template. Press **"Enter"** key after each entry in order to save all entries.



Sample No.	Cup No.	Sample ID	Sample Type	Reps	ADF	Trigger Off	MDF	Weight	Units
1	S1	TDP 0.5	Calibration Standard	2	☐	☑	☐	☐	
2	S2	TDP 0.2	Calibration Standard	2	☐	☑	☐	☐	
3	S3	TDP 0.1	Calibration Standard	2	☐	☑	☐	☐	
4	S4	TDP 0.05	Calibration Standard	2	☐	☑	☐	☐	
5	S5	TDP 0.02	Calibration Standard	2	☐	☑	☐	☐	
6	S6	TDP 0.01	Calibration Standard	2	☐	☑	☐	☐	
7	S7	TDP 0.0	Calibration Standard	2	☐	☑	☐	☐	
8	1	Blank	Unspiked	1	☐	☑	☐	☐	
9	2	Blank Spike	Spiked	1	☐	☑	☐	☐	
10	3	BLK	Unknown	1	☐	☐	☐	☐	
11	4	QC	Unknown	1	☐	☐	☐	☐	
12	5	BLK	Unknown	1	☐	☐	☐	☐	
13	6		Unknown	1	☐	☐	☐	☐	
14	7		Unknown	1	☐	☐	☐	☐	
15	8		Unknown	1	☐	☐	☐	☐	
16	9		Unknown	1	☐	☐	☐	☐	
17	10		Unknown	1	☐	☐	☐	☐	
18	11		Unknown	1	☐	☐	☐	☐	
19	12		Unknown	1	☐	☐	☐	☐	
20	13		Unknown	1	☐	☐	☐	☐	
21	14		Unknown	1	☐	☐	☐	☐	
22	15		Duplicate 1	1	☐	☑	☐	☐	
23	16		Duplicate 2	1	☐	☑	☐	☐	
24	17		Unspiked	1	☐	☑	☐	☐	
25	18		Spiked	1	☐	☑	☐	☐	
26	S16	Chk Std	Check Standard	1	☐	☑	☐	☐	
27	20		Unknown	1	☐	☐	☐	☐	
28	21		Unknown	1	☐	☐	☐	☐	
29	22	QC	Unknown	1	☐	☐	☐	☐	
30	23	BLK	Unknown	1	☐	☐	☐	☐	
31	S16	Chk Std	Check Standard	1	☐	☑	☐	☐	

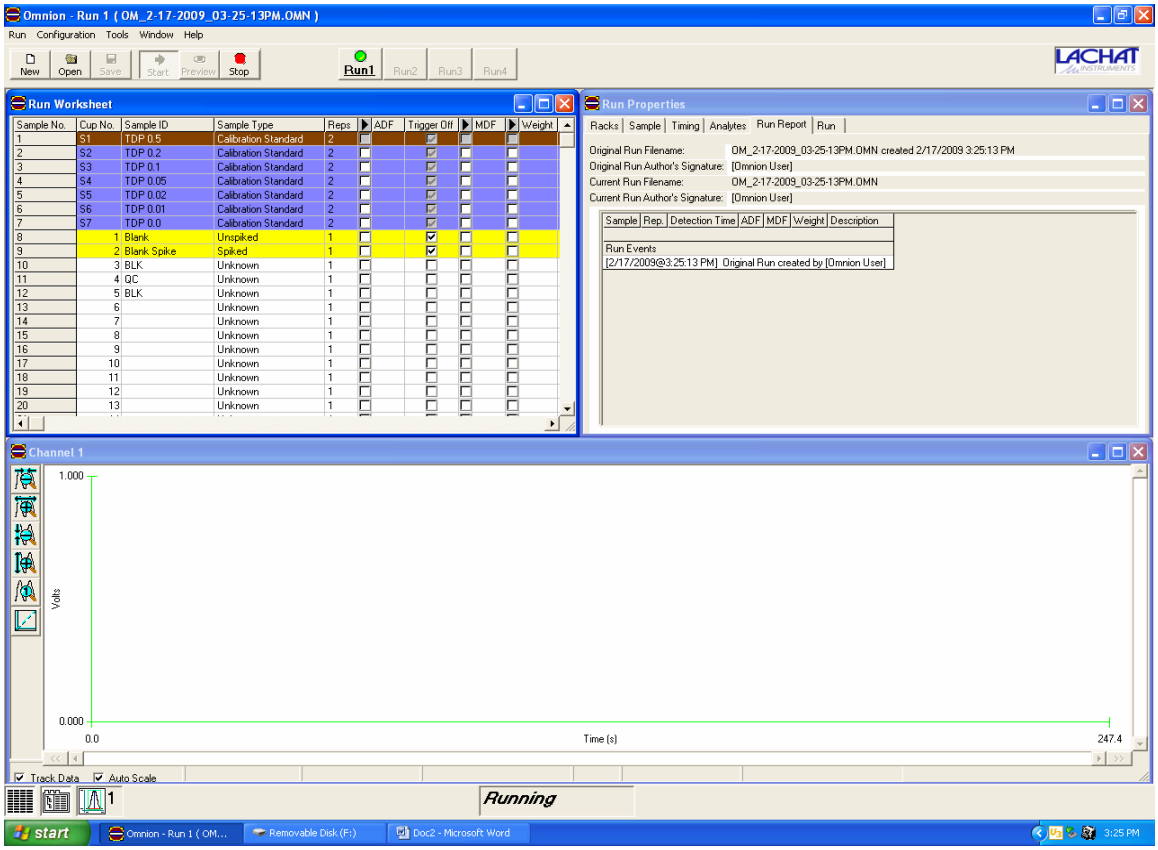
9.3.5 Print a copy of this worksheet by first double clicking on **"Run"** icon and then selecting **"Export Worksheet Data"**.

9.3.6 Click on **"Window"** tab and then, click on **"Tile"** to return to the screen with three windows.

9.3.7 Place standards in standard vials in the standard rack in order of decreasing concentration in positions 1 to 7 (std 7 is 0.00 mg/L). Place the check standard vial in position 16 (S16) of the standard rack. Pour samples and quality control samples into 13 x 100 mm test tubes and

place them in the sample racks according to the worksheet prepared in 9.2.4.

9.3.8 Pump deionized water through all reagent lines for 15 – 20 minutes and check for leaks and smooth flow. Switch to reagents and continue pumping for about 10 minutes. Click on **"Preview"** tab to monitor the baseline.



9.3.9 Once a stable baseline is achieved, click on **"Stop"** tab to stop monitoring the baseline. Click on **"Start"** tab to begin the analysis.

9.3.10 If the calibration passes, instrument will continue to analyze the samples. If it fails, take appropriate corrective actions and recalibrate before proceeding to analyze samples.

9.3.11 Manual dilution will be performed to reanalyze samples with concentration exceeding the calibrated range.

9.3.12 After the run is complete, remove the reagent lines and place them in DI water and rinse for about 15 minutes. If necessary,

rinse the system with the NaOH – EDTA rinse solution (6.1.5) for about 5 minutes followed by DI water for 10 – 15 minutes. Pump the tubes dry, release the pump tubing, and turn off the pump.

10.0 DATA ANALYSIS AND CALCULATIONS

10.1 All the calculations are performed by the Omnion 3.0 software system. It uses a calibration graph of peak area versus concentration orthophosphate concentrations in the samples. All standards are analyzed in duplicate and all data points are used for the calibration curve. Samples with orthophosphate concentrations greater than 0.5 ppm are manually diluted and reanalyzed.

10.2 Calculate the percentage spike recovery of the laboratory fortified samples as follows:

$$\% \text{ SR} = \frac{(\text{spiked sample conc.} - \text{sample conc.}),\ \text{ppm}}{\text{amount of spike added to sample},\ \text{ppm}} \times 100$$

10.3 Calculate the relative percentage difference for the duplicated samples as follows:

$$\% \text{ RPD} = \frac{\text{difference between the duplicates}}{\text{average of the duplicates}} \times 100$$

10.4 The reporting level for this method is the concentration of the lowest standard, which is 0.01 ppm.

10.5 Report results for all routine samples to three decimal places and for performance evaluation (PE) samples to three significant figures.

11.0 DATA AND RECORDS MANAGEMENT

11.1 The analysis data for the calibration blank, external QC and instrument performance check solution (IPC), i.e. the mid-range check standard, is kept on file with the sample analysis data.

11.2 Normal turnaround time for samples submitted to this lab is 2 to 10 days from the receipt. Results are reported in writing on the sample analysis request form.

11.3 The Division shall retain all pertinent laboratory records for each sample for a period of five years. At the conclusion of this period, clients shall be

given the option to take custody of their sample records. The Division shall not be responsible for retrieving, copying, or transferring laboratory records exceeding the five years custody period.

12.0 WASTE MANAGEMENT

12.1 It is laboratory's responsibility to comply with all state, federal and local regulations governing waste management, particularly the hazardous waste identification rules and disposal restrictions, and to protect the air, water and land by minimizing and controlling all releases from fume and bench operation.

12.2 Samples and standards are poured down the drain and flushed with running water.

13.0 REFERENCES

13.1 EPA Method 365.1, *Methods for the Determination of Inorganic Substances in Environmental Samples*, August 1993.

13.2 American Public Health Association, *Standard Methods for the Examination of Water and Wastewater,* 21st Edition, Method 4500- P E, 2005.

13.3 Lachat Instruments QuickChem Method 30-115-01-4-A, *Determination of Total Phosphate by Flow Injection Analysis.*

13.4 Lachat Instruments, *Operating Manual for the Quikchem Automated Ion Analyzer*.

13.5 Environmental Chemistry, DHMH-Laboratories Administration, *Quality Assurance Plan,* Revision 14.0, October 2014

13.6 Division of Environmental Chemistry, DHMH-Laboratories Administration, *Quality Manual,* Revision 1.0, November 2014

APPENDIX A
Division of Environmental Chemistry
INORGANICS ANALYTICAL LABORATORY

Data Review Checklist LL Total Dissolved Phosphorus (TDP)/Alkaline Persulfate Digestion

EPA Method 365.1

Lab Numbers: [1] _____ **Analyst:** _____

_____ **Date Digested:** _____

Dates Collected: _____ **Date Analyzed** _____

Procedure	Acceptance Criteria	Status (✓)	Comments
Holding Time	48 hours @ 4ºC 28 days @ –20ºC		
Samples Analyzed	Within 5 working days		
Calibration Curve	Corr. Coeff. ≥ 0.9950		
Reagent Blank	< Reporting level (0.010 ppm)		
Blank Spike	1 per batch		
	Recovery = 90–110%		
Matrix Spike	Every 10th sample or 1/batch, if less than 10		
	Recovery = 90–110%		
External QC	Beginning and end of each run		
	Within acceptance range		
Check Standard	After every 10th sample and at the end of the run		
	Recovery = 90–110%		
Duplicates/Replicates	Every 10th sample or 1/batch, if less than 10 samples		
	RPD ≤ 10%		
Decimal Places Reported	3		
Measured Values	Within calibration range (0.010–0.500 ppm)		
Diluted Samples	Correct final calculations		
Changes/Notes	Clearly stated		

[1] Include beginning and ending numbers, account for gaps by bracketing.

_____ _____

Analyst's Signature & Date Reviewer's Signature & Date

_____

Supervisor's Signature & Date

Reagents	ID	Reagents	ID	External QC	
Color Reagent		Oxidizing Reagent		Identification =	
Ascorbic Acid		Borate Buffer		True Value =	ppm
1M HCl				Range =	ppm

Nitrate/Nitrite and Nitrite (Low Level) Flow Injection Colorimetric Analysis

EPA Method 353.2

1.0 SCOPE AND APPLICATION

1.1 This method determines nitrite, or nitrate/nitrate in drinking, ground, surface, domestic waters and industrial waste.

1.2 The range of this method is from 0.020 mg/L to 4.00 mg/L for nitrate–nitrite and 0.002 mg/L to 0.400 mg/L for nitrite.

1.3 Nitrate is found only in small amounts in domestic wastewater, but in the effluent of nitrifying biological treatments nitrate may be found in concentrations up to 30 mg/L.

2.0 SUMMARY OF METHOD

Nitrate is reduced quantitatively to nitrite by passage through a copperized cadmium column. The reduced nitrate plus original nitrite is further determined by coupling with N-(naphthyl) ethylenediamine dihydrochloride. The dye produced has magenta color and is read at 520nm, and is directly proportional to the concentration of analyte. Determination of nitrite takes place through the same procedure as Nitrate/Nitrite without a cadmium column.

3.0 INTERFERENCES

3.1 Interference from iron, copper or other metals is eliminated by addition of EDTA to the ammonium chloride buffer.

3.2 Suspended matter in the column will restrict sample flow. Remove suspended solids by filtration.

3.3 Method interferences may be caused by contaminants in the reagent water, reagents, glassware, and other sample processing apparatus that can bias analyte response.

4.0 HEALTH AND SAFETY

4.1 Accepted laboratory safety practices should be followed during reagent preparation and instrument operation. Use of lab coats, fume hoods, gloves and eye protection are required.

4.2 The following chemicals have the potential to be highly toxic or hazardous.

4.2.1 Cadmium
4.2.2 Phosphoric acid

4.2.3 Hydrochloric acid

4.2.4 Sodium Hydroxide

4.2.5 A reference file of Material Safety Data Sheet (MSDS) is available to all personnel involved in the chemical analysis.

5.0 EQUIPMENT AND SUPPLIES

5.1 Equipment

5.1.1 Lachat QuickChem 8500 Automated Ion Analyzer, which includes the following:

5.1.1.1 Automatic sampler

5.1.1.2 Multi-channel proportioning pump

5.1.1.3 Injection module with a 12.5 cm x 0.5 mm ID sample loop

5.1.1.4 Manifold

5.1.1.5 Colorimetric detector

5.1.1.5.1 Flow cell, 10 mm path length

5.1.1.5.2 Interference filter, 520 nm

5.1.1.6 Computer with Omnion 3.0 Data system and printer

5.1.2 Analytical balance capable of weighing to the nearest 0.0001 g

5.1.3 Top loading balance for weighing chemicals for reagents

5.2 Supplies

5.2.1 Class A volumetric flasks, 50 – 1,000 mL

5.2.2 Class A volumetric pipettes, 1– 10 mL

5.2.3 Automatic pipetters, 100 µL- 10 mL

5.2.4 Beakers, disposable, polypropylene, 50 mL(Fisher 01-291-10)

5.2.5 Test tubes, glass, 13 x 100 mm and 16 X 125 mm

5.2.6 Reagent storage bottles, plastic or glass

5.2.7 Cadmium Reduction Column (Lachat Part No. 50237A)

5.2.8 Ultra High Purity Helium gas for degassing

6.0 REAGENTS AND STANDARDS

6.1 Reagents

6.1.1 Ammonium Chloride buffer, pH 8.5, 2 L - Dissolve 170 g of NH_4Cl and 2.0 g of disodium ETDA ($Na_2EDTA'2H_2O$) in about 1600 mL DI water in a 2 L beaker. Mix using a stir bar. Adjust the pH to 8.5 with 15 N sodium hydroxide solution and bring up to volume. Use filter paper to remove all the small particles from the reagent and refrigerate. Prepare monthly.

6.2.1 Sulfanilamide color Reagent, 1 L - Add carefully, while mixing, 100 mL 85% phosphoric acid (H_3PO_4) to 700 mL DI water in a 1 L volumetric flask. Add 40 mg sulfanilamide and 1g N-1-naphthyl ethylenediamine dihydrochloride (NED) and stir to dissolve. Bring up to 1 L with DI water. Filter, store in amber bottle and refrigerate. This solution is stable for one month.

6.2.1 15 N Sodium Hydroxide – Add 150 g NaOH very slowly to 180 mL DI water in a 250 mL volumetric flask. CAUTION: The solution will get very hot! Mix until dissolved. Cool and store in a *plastic* bottle.

6.2 Standards

6.2.1 Nitrate Stock Standard (1000 mg /L of nitrate nitrogen) - Purchased from approved commercial supplier with expiration date. If this standard is not available, then weigh 0.7218g of dried potassium nitrate KNO_3 (1000 mg/ L of nitrate nitrogen) in 100 mL volumetric flask

.

6.2.2 Nitrite Stock Standard (1000 mg/L of nitrite nitrogen) - Purchased from approved commercial supplier with expiration date. If this standard is not available, then weigh 0.6072g of dried potassium nitrite KNO_2 (1000 mg/L of nitrite nitrogen) in 100 mL volumetric flask.

6.2.3 Combined Intermediate Standard, 90 mg/L nitrate nitrogen and 10 mg/L nitrite nitrogen - Pipete 9 mL of 6.2.1 and 1 mL of 6.2.2 into about 70 mL DI water in a 100 mL volumetric flask. Bring up to volume with DI water, mix, and store at 4^{o}C. Use this standard as spiking solution.

6.2.4 Nitrate Cadmium check, 0.5 ppm – Dilute 100 µL of reagent 6.2.1 to 200 mL with DI water in a 200 mL volumetric flask.

6.2.5 Nitrite Cadmium check, 0.5 ppm – Dilute 100 µL of reagent 6.2.2 to 200 mL with DI water in a 200 mL volumetric flask.

6.2.6 Working Standards - The working standards are prepared by diluting the combined intermediate standard (6.2.3) in 100 mL volumetric flasks according to the following table once every two days:

NO3+NO2 ppm	NO2 ppm	Combined Intermediate Std
4.000	0.400	4 mL

2.000	0.200	2 mL
1.000	0.100	1 mL
0.500	0.050	500 µL
0.200	0.020	200 µL
0.080	0.008	80 uL
0.020	0.002	20 uL
0.000	0.000	DI H_2O

7.0 SAMPLE COLLECTION, PRESERVATION, AND STORAGE

7.1 Samples are collected in plastic bottles or cubitainers, and are preserved by cooling to 4^{o} C. These samples are never acidified and their pH is usually between 5 to 9. Adjust the pH with Conc. HCl or Conc. NH_4OH, if the pH is below 5 or above 9.

7.2 Samples are analyzed within 48 hours after collection. If they cannot be analyzed within this time period, they should be frozen at -20^{o}C on the first day they arrive to lab. The holding time for frozen samples is 28 days.

8.0 QUALITY CONTROL

8.1 An initial Demonstration of Performance (IDP) to establish the instrument's capability is required prior to its use in analyzing client's samples. The linearity of calibration range (LCR) and the ability to quantify correctly Quality Control Samples are used to assess performance.

8.2 All the standards are run in duplicates: that is, two analyses from each tube. A linear calibration is performed daily before the sample run.

8.3 A mid-range check standard and a calibration blank is analyzed following daily calibration, after every ten samples (or more frequently, if required) and at the end of the sample run. If a check standard fails, the samples between the last satisfactory check standard and the unsatisfactory standard are re-analyzed.

8.4 Every tenth sample is duplicated (analyzed from two different tubes) and spiked. The accepted value for the relative percentage relative difference (RPD) and spike recovery is ± 10 %. Prepare sample spikes by adding 50 µL of 6.2.3 to 10 mL of samples. If these do not fall within the accepted ranges, the corresponding analyses are repeated.

8.5 An external quality control is analyzed at the beginning and at the end of each analytical run.

8.6 A deionized water blank is run in the beginning and after every tenth sample. Results for blanks should be <0.002 for NO2 and <0.02 for NO3+NO2 mg N/L.

8.7 A method detection limit (MDL) study is performed once a year by analyzing seven replicates of the 0.1 ppm standard spread over three analytical runs. MDL is calculated as follows:

MDL = (t) x (S) where, t = Student's t value for a 99% confidence level and a standard deviation estimate with n-1 degrees of freedom [t = 3.14 for seven replicates] and S = standard deviation of the replicate analyses.
Each new analyst also performs an MDL study or when any changes in the analytical procedure or instrument are made.

8.8 A demonstration of capability study is performed by each analyst performing the test by analyzing four replicates of an external quality control sample during one or two analytical runs.

9.0 PROCEDURE

9.1 Sample preparation

9.1.1 Prepare a list of samples to be analyzed and pour samples into labeled tubes (16 mm x 125 mm test tubes).

9.1.2 Spike the blank and every tenth sample by adding 50 µL of combined standard (6.2.3) to 10 mL of sample or DI water.

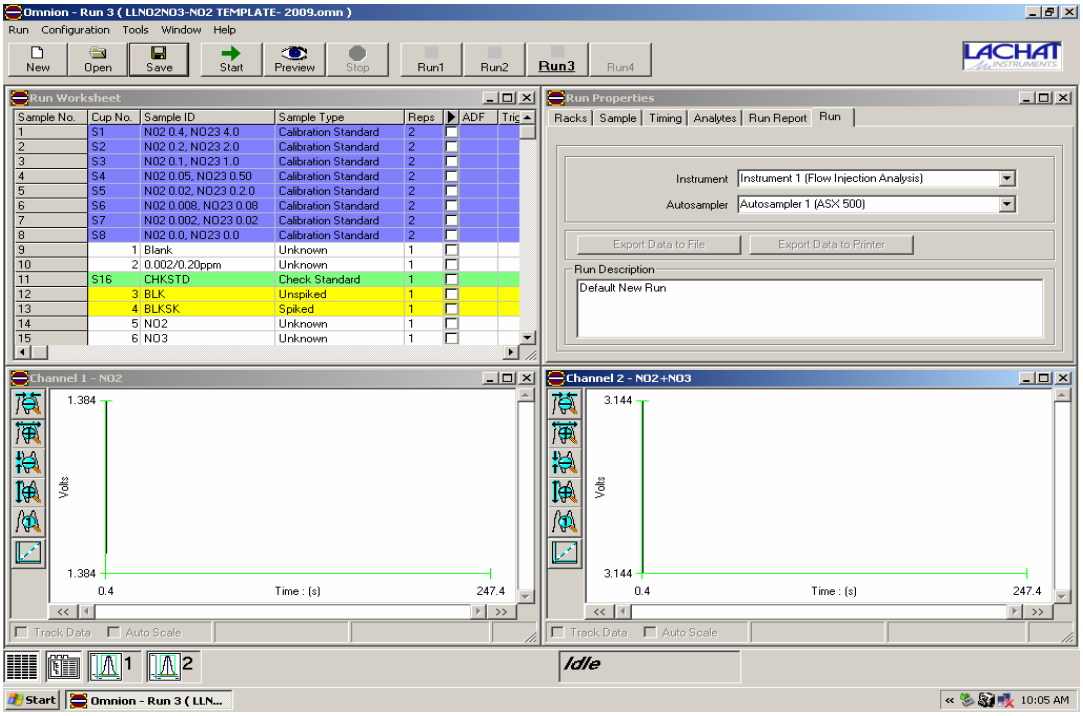
9.1.3 To prevent bubble formation, degas all reagents with helium, except those specified not to. Use helium at 140 Pa (20lb/in2)

9.2 Instrument calibration and sample analysis

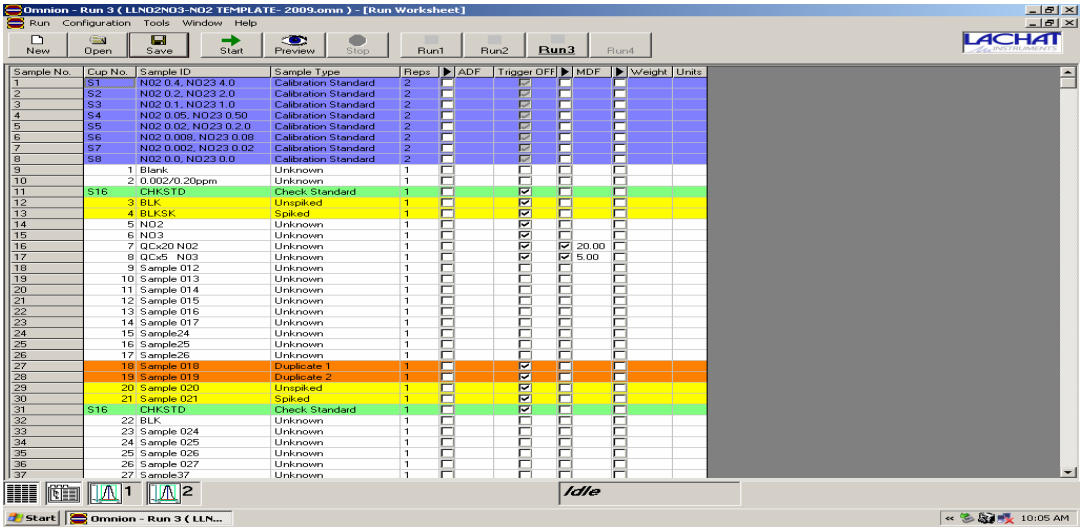
9.2.1 Set up manifold as in the method's manifold diagram.

9.2.2 Turn on the Lachat instrument, computer, monitor, and the printer.

9.2.3 Double click on Omnion and open the **"LL NO_3+NO_2/ NO_2"** folder to find the template, which consists of four windows.



9.2.4 Maximize the **"Run Worksheet"** window at the top left hand corner of the screen by clicking on the middle square on that screen.



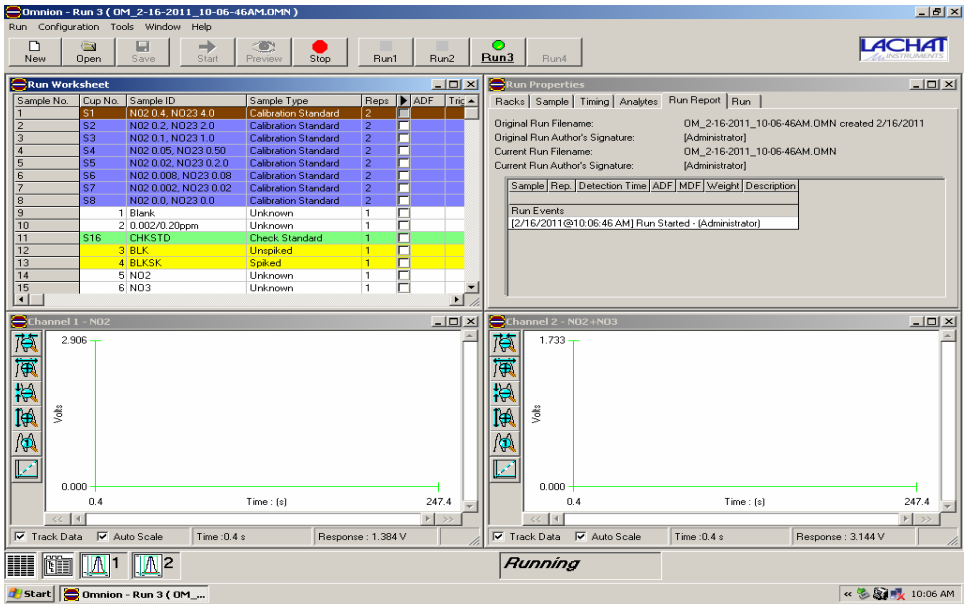
9.2.5 Enter all the sample, sample spike, QC and standard information in the run worksheet and print out the list.

9.2.6 Click on **"Window"** tab and then, click on **"Tile"** to return to the screen with three windows (9.2.3).

9.2.7 Place standards in standard vials in the standard rack in order of decreasing concentration in positions 1 to 8. Place the check standard vial in position 16 (S16) of the standard rack. Pour samples and quality control samples into 13 x 100 mm test tubes and place them in the sample racks according to **"Sample Run Log"**.

9.2.8 Pump deionized water through all reagent lines for 15 – 20 minutes and check for leaks and smooth flow. Switch to reagents, turn on the Cadmium switching valve on and allow the buffer to rinse it for 5-10 minutes (Note: Lachat columns come as ready to use and need to be flushed for about 10 minutes with buffer after installing on the system.) Click on **"Preview"** tab to monitor the baseline.

9.2.9 Once a stable baseline is achieved, click on **"Stop"** tab to stop monitoring the baseline. Click on **"Start"** tab to begin the analysis.



9.2.10 If the calibration passes, instrument will continue to analyze the samples. If it fails, take appropriate corrective actions and recalibrate before proceeding to analyze samples.

9.2.11 Auto dilution will trigger on to reanalyzed samples with concentration exceeding the calibrated range.

9.2.12 When the run is complete, remove the reagent lines and place them in DI water and rinse for about 15 minutes. For extra rinse a reagent of Disodium EDTA can be used followed by DI rinse. Then all the reagent lines should be air dried and released from the pump.

10.0 DATA ANALYSIS AND CALCULATIONS

10.1 All the calculations are performed by the Omnion 3.0 software system. It uses a calibration graph of peak area versus concentration to calculate the "nitrate + nitrite nitrogen" concentrations in the samples. All standards are analyzed in duplicate and all data points are used for the calibration curve. Samples with

"nitrate + nitrite nitrogen" concentrations greater than 4 ppm are automatically diluted and reanalyzed.

10.2 The reduction efficiency of the cadmium coil is calculated as followings:

$$\frac{NO_3 \text{ (peak height)}}{NO_2 \text{ (peak height)}} \text{ X } 100$$

10.3 Calculate % of spike recovery of the laboratory fortified samples as follows:

$$\% \text{ SR} = \frac{\text{spiked sample conc. - sample conc., ppm}}{\text{amount of spike added to sample, ppm}} \text{ x } 100$$

10.4 Calculate the relative percent difference for the duplicated samples as follows:

$$\text{RPD} = \frac{\text{difference between the two duplicates}}{\text{average of the duplicates}} \text{ x } 100$$

11.0 DATA AND RECORDS MANAGEMENT

11.1 All the results are reported on the sample request forms to three decimal places and for performance evaluation (PE) samples to three significant figures. Normal turnaround time for samples submitted to this lab is 2 to 10 days from receipt.

11.2 Completed data packages are scanned and stored electronically before being placed in the appropriate binders in the lab.

11.3 Results are reported in writing on a sample analysis request form. The Division shall retain all pertinent laboratory records for each sample for a period of five years. At the conclusion of this period, clients shall be given the option to take custody of their sample records. The Division shall not be responsible for retrieving, copying, or transferring laboratory records exceeding the five years custody period.

12.0 WASTE MANAGEMENT

12.1 It is the laboratory's responsibility to comply with all state, federal and local regulations governing waste management, particularly the hazardous waste identification rules and disposal restrictions, and to protect the air, water and land by minimizing and controlling all releases from fume and bench operation.

12.2 Samples and standards are poured down the drain while large amount of water is running.

12.3 Compliance with state's sewage discharge permits and regulations is required. For more information consult the "Waste Management manual for Laboratory Personnel", available from the American Chemical Society's Department of Government Regulations and Science Policy, 1155 16th Street N. W., Washington D. C. 20036, (202) 872-4600.

13.0 REFERENCES

13. EPA Method 353.2, *Methods for the Determination of Inorganic Substances in Environmental Samples,* Revision 2.0*,* August 1993.

13.2 American Public Health Association, *Standard Methods for the Examination of Water and Wastewater, 21st Edition, p. 4-125, Method 4500- NO_3^-, 2005*

13.3 Lachat Instruments, Methods Manual for the *Quikchem Automated Ion Analyzer*, Method 10-107-04-1-A.

13.4 Division of Environmental Chemistry, DHMH-Laboratories Administration, *Quality Assurance Plan,* Revision 14.0, October 2014

13.6 Division of Environmental Chemistry, DHMH-Laboratories Administration, *Quality Manual,* Revision 1.0, November 2014

APPENDIX **A**

Division of Environmental Chemistry
INORGANICS ANALYTICAL LABORATORY

Data Review Checklist – Nitrate + Nitrite Nitrogen (Low Level)

EPA Method 353.2; Revision 3.0

Lab Numbers: [1] _____ **Analyst:** _____

Dates Collected: _____ **Date Analyzed:** _____

Procedure	Acceptance Criteria	Status (√)	Comments
Holding Time	48 hours @ 4ºC 28 days @ –20ºC		
Calibration Curve	Corr. Coefficient. ≥ 0.9950		
Reagent Blank	< 0.02 ppm for NO23 and < 0.002 ppm for NO2		
Blank Spike	1 per batch		
	Recovery = 90 – 110%		
Matrix Spike	Every 10th sample or 1/batch, if less than 10 samples		
	Recovery = 90–110%		
External QC	Beginning and end of each run		
	Within acceptable range		
Check Standard	After every 10th sample and at the end of the run		
	Concentrations = 90–110% of the true value		
Duplicates/Replicates	Every 10th sample or 1/batch, if less than 10 samples		
	RPD ≤ 10%		
NO_3/NO_2 Cadmium Column Check	90–110%		
Decimal Places Reported	3		
Measured Values	Within calibration range (0.020–4.000 ppm for NO_2+NO_3; 0.002–0.400 ppm for NO_2)		
Diluted Samples	Correct final calculations		
Changes/Notes	Clearly stated		

***** Check (√) if criteria are met. [1]Include beginning and ending numbers, account for gaps by bracketing

_____ _____
Analyst's Signature & Date Reviewer's Signature & Date

_____
Supervisor's Signature & Date

Reagents	ID
Ammonia Buffer	
Color Reagent	

External QC

Identification = _____

True Value =	NO2	NO2+3	ppm
Range =	NO2	NO2+3	ppm

DETERMINATION OF AMMONIA (LOW LEVEL) FLOW INJECTION COLORIMETRIC ANALYSIS

EPA Method 350.1

1.0 SCOPE AND APPLICATION

1.1 This method determines Ammonia in industrial samples, drinking, ground and surface waters.

1.2 The applicable range of this method is 0.008 to 0.500 mg N/L.

2.0 SUMMARY OF METHOD

This method is based on the Berthelot reaction. In the reaction of ammonia with alkaline phenol and sodium hypochlorite, an indophenol blue is formed. Sodium nitroprusside is added to enhance sensitivity. The absorbance of the reaction product is measured at 630 nm, and is directly proportional to the ammonia concentration in the sample.

3.0 INTERFERENCES

3.1 Sufficient concentration of calcium and magnesium ions can precipitate if they are present in sufficient concentrations. Tartrate or EDTA addition to the sample can prevent this problem.

3.2 Color, turbidity and certain organic species may interfere. Turbidity is removed by filtration and sample color can be corrected for by running the samples through the manifold without color formation.

3.3 Method interferences may be caused by contaminants in the reagent water, reagents, glassware, and other sample processing apparatus that can bias analyte response, especially in low level detection of Ammonia. To eliminate this problem, wash glassware with 1:1 HCl and rinse with DI water.

4.0 HEALTH AND SAFETY

4.1 Accepted laboratory safety practices should be followed during reagent preparation and instrument operation. Use of gloves, eye protection, and lab coat are required when preparing reagents.

4.2 The following chemicals have the potential to be highly toxic or hazardous.

4.2.1 Phenol
4.2.2 Sodium nitroprusside

4.3 A reference file of Material Safety Data Sheet (MSDS) is available to all personnel involved in the chemical analysis.

5.0 EQUIPMENT AND SUPPLIES

5.1 Equipment

5.1.1 Lachat Quick Chem FIA 8500 series.

5.1.1.2 XYZ Auto sampler ASX-520 series with sample, standard and dilution racks

5.1.1.3 Manifold or reaction unit

5.1.1.4 Multichannel Reagent Pump RP-100 series

5.1.1.5 Colorimetric Detector

5.1.1.5.1 Flowcell, 10 mm, 80uL, glass flow cell

5.1.1.5.2 630 nm interference filter

5.1.1.6 Computer, monitor, printer and The Flow Solution software.

5.2 Supplies

5.2.1 13x100 mm test tubes, Fisher # 14-961-27

5.2.2 16x125 mm test tubes, Fisher # 14-961-30

6.0 REAGENTS AND STANDARDS

6.1 Reagents

6.1.1 Sodium Phenolate- In a 1 L volumetric flask, dissolve 88 ml of 88% liquefied phenol. While stirring, slowly add 32 g sodium hydroxide (NaOH). Cool and invert to mix. Do not degas this reagent. Prepare fresh every 3 to 5 days and save in amber container. Discard when the reagent turns brown. Always prepare this reagent under the hood.

6.1.2 Sodium Nitroprusside- In a 1 L volumetric flask, dissolve 3.5 g sodium nitroprusside. Mix and dilute to the mark with DI water. Prepare fresh every 1 to 2 weeks.

6.1.3 Hypochlorite Reagent - To a 500 mL volumetric flask, add 250 mL sodium hypochlorite (NaOCl-5.25%) and bring up to volume with DI water. If using the 6% sodium hypochlorite, add 219 mL of bleach and bring up to volume.

6.1.4 1 M Sodium Hydroxide Solution- In a 1 L volumetric flask, dissolve 40.0 g sodium hydroxide in approximately 900 ml DI water. Dilute to the mark after all is dissolved.

6.1.5 Buffer for non acidified samples- In a 1 L volumetric flask, dissolve 50.0g disodium ethylenediamine tetraacetic acid (Na2EDTA) and 225 ml 1 M sodium hydroxide in approximately 700 ml DI water. Mix well and dilute to the mark. Prepare fresh monthly.

6.1.6

6.1.7

Ammonia ppm	Combined Intermediate Std	Final Volume
0.000	DI water	100 ml
0.008	2.67 ml of Std 0.30ppm	100 ml
0.020	20 ml std 0.100ppm	100 ml
0.100	100 µl	100 ml
0.200	200 µl	100 ml
0.300	600 µl	200 ml
0.400	400 µl	100 ml
0.500	500 µl	100 ml

6.2

Standards

6.2.1 Ammonia Stock Standard (1000) mg N/L - This standard is pre-made and purchased from an approved commercial supplier with expiration date. If this stock standard is not available, prepare it by dissolving 0.3819 g ammonium chloride (NH_4Cl) that has been dried in the oven for two hours at 105 ^{o} C, in about 80 ml of DI water. Bring up to the 100 mL mark with DI water and store at 4^{o} C. Prepare this reagent monthly.

6.2.2 Intermediate Standard (100 mg N/L) - Pipette 10 ml of standard 6.2.1 into a 100 ml volumetric flask. Bring up to mark with DI water. Store at 4^{o} C. Make weekly.

6.2.3 Spiking Solution (100 mg N/L) - This is the same as the intermediate standard, which is used to spike the samples. Pipette 30 uL of the spiking solution (standard 6.2.2) into 10 mL of DI water or 10 mL of sample, in order to make the blank spike and sample spike. The concentration of spiking solution is 0.30 mg/L

6.2.4 Working Standards - The working standards are prepared weekly according to the following table:

7.0 SAMPLE COLLECTION, PRESERVATION, AND STORAGE

7.1 Samples are collected in plastic or glass bottles. All bottles must be thoroughly cleaned and rinsed with reagent water.

7.2 Never use acid preservation for samples to be analyzed for Low Level Ammonia.

7.3 Samples to be analyzed for ammonia only are cooled to 4^{o} C and analyzed within 48 hours. For short-term preservation, freeze at –20^{o} C.

8.0 QUALITY CONTROL

8.1 An Initial Demonstration of Performance (IDP) to establish the instrument's capability is required prior to its use in analyzing client's samples. The Linearity of Calibration Range (LCR) and the ability to quantify the Quality Control Samples correctly are used to assess performance.

8.2 A demonstration of capability study is performed by each analyst performing the test by analyzing four replicates of an external quality control sample during one or two analytical runs.

8.3 All the standards are run in duplicates: that is, two analyses from each tube. A linear calibration is done daily before the sample run. See the attached checklist for the acceptance criteria.

8.4 A mid-range check standard and a calibration blank is analyzed immediately following daily calibration, after every ten samples (or more frequently, if required) and at the end of run. If a check standard fails, the samples between the last satisfactory check standard and the unsatisfactory standard are re-analyzed.

8.5 Every tenth sample is duplicated (analyzed from two different tubes) and spiked. The accepted value for the relative percent difference (RPD) or spike recovery is ± 10 %. If these do not fall within the accepted ranges, the corresponding analyses are repeated.

8.6 A known QC is analyzed for ammonia in the beginning and at the end of each run.

8.7 A method detection limit (MDL) study is performed once a year by analyzing seven replicates of the 0.008 ppm standard spread over three analytical runs. MDL is calculated as follows:

MDL = (t) x (S) where, t = Student's t value for a 99% confidence level and a standard deviation estimate with n-1 degrees of freedom [t = 3.14 for seven replicates] and S = standard deviation of the replicate analyses.
Each new analyst also performs an MDL study or when any changes in the analytical procedure or instrument are made.

9.0 PROCEDURE

9.1 Sample preparation

9.1.1 Prepare a list of samples to be analyzed and pour samples into labeled tubes (16mm x 125 mm test tubes).

9.1.2 Spike every tenth sample by adding 30 uL of 100 ppm N/L (Intermediate standard) into 10 mL DI water (blank spike) or 10 mL of sample (sample spike).

9.1.3 Filter the turbid samples by inserting the Sera Filter inside the 16 X 125 mm test tubes containing the sample. Press the filter down and pour the filtered sample collected on the top inside a 13 x 100 mm test tube for analysis.

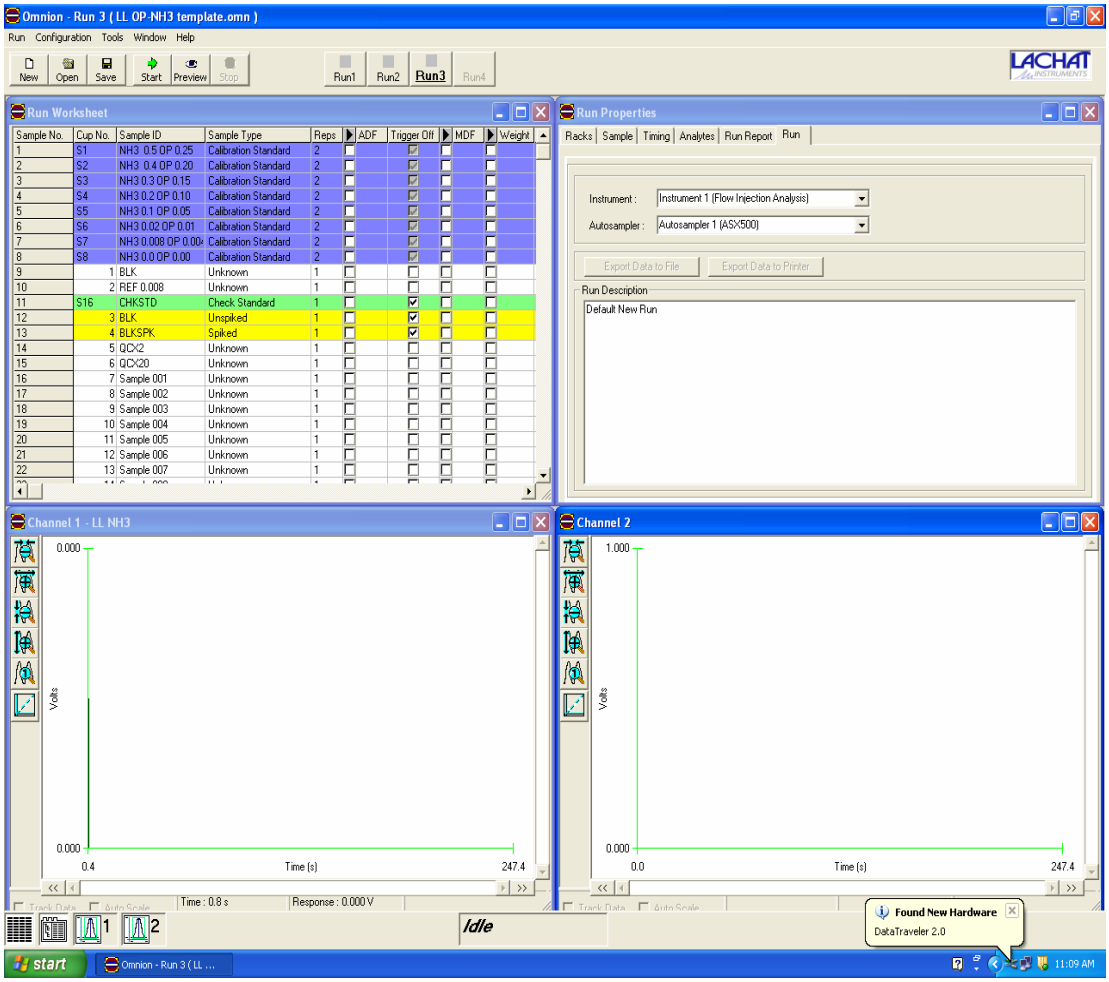
9.1.4 To prevent bubble formation, degas all reagents, except for phenol, with helium for two minutes. Use Helium at 140 kPa (20lb/in2) through a helium degassing tube or a pipette.

9.2 Instrument set-up and sample analysis

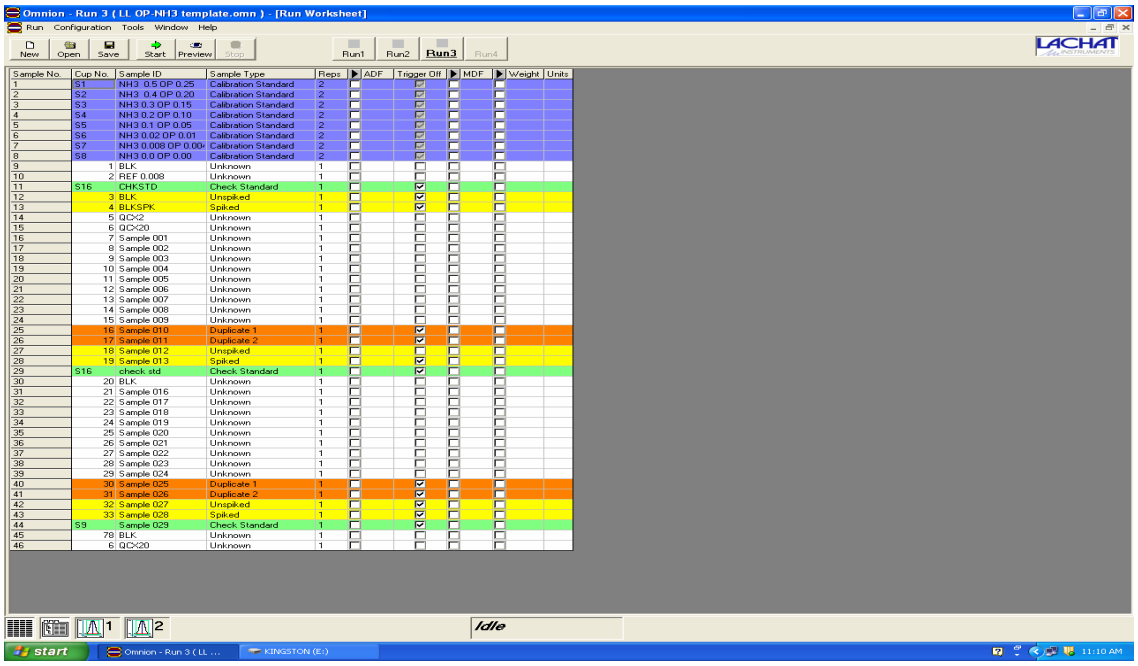
9.2.1 Set up manifold as in the diagram.

9.2.2 Turn on the Lachat instrument, computer, monitor, and printer.

9.2.3 Double click on **"LL OP/NH3"** to open the template, which consists of four windows. Samples are analyzed consecutively for orthophosphate and ammonia on the same system.



9.2.4 Maximize the **"Run Worksheet"** window at the top left hand corner of the screen, by clicking on the middle square on that screen to open the worksheet template. Enter sample identifications in the sample ID column, making sure that all duplicates and spikes are entered in the appropriate locations of the worksheet. Identifications of the quality control samples analyzed at the beginning of the run and the locations of the duplicates and the spiked have been saved on the template. Press **"Enter"** key after each entry in order to save all entries.

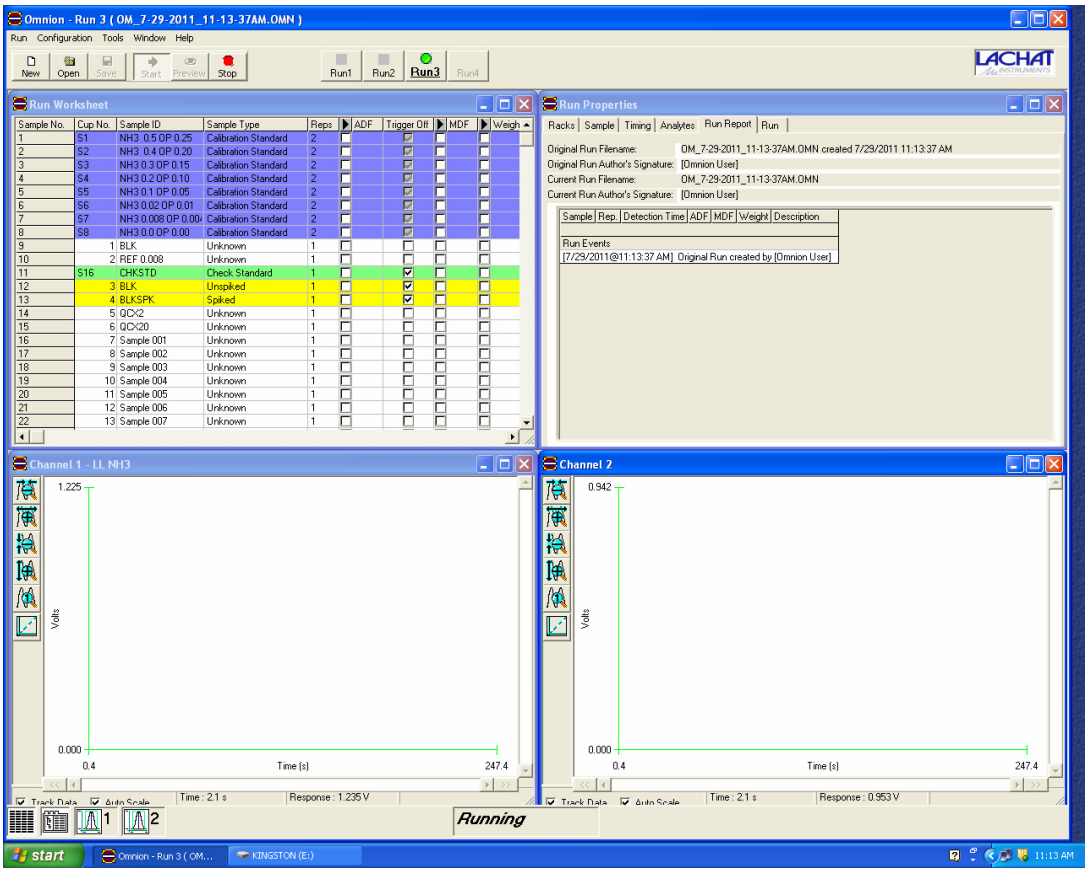


9.3.5 Print a copy of this worksheet by first double clicking on **"Run"** icon and then selecting **"Export Worksheet Data"**.

9.3.6 Click on **"Window"** tab and then, click on **"Tile"** to return to the screen with four windows.

9.3.7 Place standards in standard vials in the standard rack in order of decreasing concentration in positions 1 to 8 (STD 8 is DI water-0.00 mg/L). Place the check standard vial in position 16 (S16) of the standard rack. Pour samples and quality control samples into 13 x 100 mm test tubes and place them in the sample racks according to the worksheet prepared in 9.2.4.

9.3.8 Pump deionized water through all reagent lines for 15 – 20 minutes and check for leaks and smooth flow. Switch to reagents in the order of 1. Buffer, 2. Phenol, 3. Bleach, 4. Nitroprusside, 5. Ascorbic Acid, 6. Color Reagent and continue pumping for about 10 minutes. Click on **"Preview"** tab to monitor the baseline.



9.3.9 Once a stable baseline is achieved, click on **"Stop"** tab to stop monitoring the baseline. Click on **"Start"** tab to begin the analysis.

9.3.10 If the calibration passes, the instrument will continue to analyze the samples. If it fails, take appropriate corrective actions and recalibrate before proceeding to analyze samples.

9.3.11 Manual dilution will be performed to reanalyze samples with concentrations exceeding the calibrated range.

9.2.12 After the run is complete, remove the reagent lines in the reverse order that they were inserted and place them in DI water and rinse for about 15 minutes. For extra rinse of NH3 channel, a reagent of 1M HCl can be used for 5 minutes followed by DI rinse for 10 – 15 minutes. Pump the tubes dry, release the pump tubing, and turn off the pump.

10.0 DATA ANALYSIS AND CALCULATIONS

10.1 All the calculations are performed by the Omnion 3.0 software system. It uses a calibration graph of peak area versus concentration to calculate the ammonia nitrogen concentrations in the samples. All standards are analyzed in duplicate and all data points are used for the calibration curve. Samples with nitrogen concentrations greater than 0.500 ppm are manually diluted and reanalyzed.

10.2 Calculate the percentage spike recovery of the laboratory fortified samples as follows:

$$\%\,SR = \frac{(\text{spiked sample conc.} - \text{sample conc.}),\ \text{ppm}}{\text{amount of spike added to sample, ppm}} \times 100$$

10.3 Calculate the relative percentage difference for the duplicated samples as follows:

$$\%\,RPD = \frac{\text{difference between the duplicates}}{\text{average of the duplicates}} \times 100$$

11.0 DATA AND RECORDS MANAGEMENT

11.1 Completed data packages are scanned and stored electronically before being placed in the appropriate binders in the lab.

11.2 Report only those results that fall between the lowest and the highest calibration standards. Samples exceeding the highest standard are manually diluted and reanalyzed.

11.3 Sample results for are reported in mg N/L. Report results to three decimal places. Report results below the lowest calibration standard as <0.008. For the Chesapeake Bay Program only, report all calculated results with the "L" sign for concentrations less than that of the lowest Standard.

12.0 WASTE MANAGEMENT

12.1 It is laboratory's responsibility to comply with all state, federal and local regulations governing waste management, particularly the hazardous waste identification rules and disposal restrictions, and to protect the air, water and land buy minimizing and controlling all releases from fume and bench operation.

12.2 Samples and standards are poured down the drain while a large amount of water is running. Ammonia waste containing phenol is collected in a waste drum under the hood and handled according to the laboratory and state's regulations. For more information consult the "Waste Management Manual for Laboratory Personnel", available from the American Chemical Society's Department of Government Regulations and Science Policy, 1155 Street N. W., Washington D. C. 20036, (202) 872-4477.

13.0 REFERENCES

13.1 EPA Method 350.1, *Methods for the Determination of Inorganic Substances in Environmental Samples*, August 1993

13.2 American Public Health Association, *Standard Methods for the Examination of Water and Wastewater,* 21st Edition, Method 4500- NH3 H, 2005

13.3 *Lachat Instruments QuickChem Method 10 – 107 – 06 – 1 – J,* Determination of Ammonia by Flow Injection Analysis

13.4 Lachat Instruments, *Operating Manual for the Quikchem Automated Ion Analyzer*

13.5 Division of Environmental Chemistry, DHMH-Laboratories Administration, *Quality Assurance Plan,* Revision 14.0, October 2014

13.6 Division of Environmental Chemistry, DHMH-Laboratories Administration, *Quality Manual,* Revision 1.0, November 2014

APPENDIX A

Division of Environmental Chemistry
INORGANICS ANALYTICAL LABORATORY

Data Review Checklist-LL Ammonia

EPA Method 350.1

Lab Numbers: [1] _____ **Analyst:** _____

Dates Collected: _____ **Date Analyzed:** _____

Procedure	Acceptance Criteria	Status(√)	Comments
Holding Time	48 hours @ 4°C 28 days @ –20°C		
Calibration Curve	Corr. Coeff. ≥ 0.9950		
Reagent Blank	< Reporting level (0.008 ppm)		
Blank Spike	1 per batch		
	Recovery = 90–110%		
Matrix Spike	Every 10th sample or 1/batch, if less than 10 samples		
	Recovery = 90–110%		
External QC	Beginning and end of each run		
	Within acceptable range		
Check Standard	After every 10th sample and at the end of the run		
	Concentration = 90–110% of the true value		
Duplicates/Replicates	Every 10th sample or 1/batch, if less than 10 samples		
	RPD ≤ 10%		
Decimal Places Reported	3		
Measured Values	Within calibration range (0.008–0.500 ppm)		
Diluted Samples	Correct final calculations		
Changes/Notes	Clearly stated		

[1] Include beginning and ending numbers, account for gaps by bracketing.

_____ _____

Analyst's Signature & Date Reviewer's Signature & Date

_____

Supervisor's Signature and Date

Reagents	ID	Reagents	ID
Sodium Phenolate		Sodium Hypochlorite	
Sodium Nitroprusside		EDTA Buffer	

External QC

Identification = _____

True Value = _____ ppm

Range = _____ ppm

APPENDIX B

\Division of Environmental Chemistry
INORGANICS ANALYTICAL LABORATORY

Data Review Checklist LL Orthophosphate/ LL Ammonia

EPA Method 365.1/ EPA Method 350.1

Lab Numbers: [1] _____

Analyst: _____

Dates Collected: _____ **Date Analyzed:** _____

Procedure	Acceptance Criteria	Status(√)	Comments
Holding Time	48 hours @ 4°C; 28 days @ –20°C		
Calibration Curve	Corr. Coeff. ≥ 0.9950		
Reagent Blank	< Reporting Level (0.004 ppm for OP; 0.008 ppm for NH_3)		
Blank Spike	1 per batch		
	Recovery = 90–110%		
Matrix Spike	Every 10th sample or 1/batch, if less than 10		
	Recovery = 90–110%		
External QC	Beginning and end of each run		
	Within acceptable range		
Check Standard	After every 10th sample and at the end of the		
	Concentration = 90–110% of the true value		
Duplicates/Replicates	Every 10th sample or 1/batch, if less than 10		
	RPD ≤ 10%		
Decimal Places Reported	3		
Measured Values	Within calibration range (0.004–0.250 ppm for OP; 0.008–0.500 ppm for NH_3)		
Diluted Samples	Correct final calculations		
Changes/Notes	Clearly stated		

[1] Include beginning and ending numbers, account for gaps by bracketing.

_____ _____
Analyst's Signature & Date Reviewer's Signature & Date

_____
Supervisor's Signature and Date

NH_3 Reagents	ID	OP Reagents	ID
Sodium Phenolate		Color Reagent	
Sodium Nitroprusside		Ascorbic Acid	
Sodium Hypochlorite			
EDTA Buffer			

External QC

Identification = _____

True Value = NH_3 _____ /OP _____ ppm

NH_3 Range = _____ ppm

OP Range = _____ ppm

Standard Operating Procedure

Low Level Orthophosphate (Flow Injection Colorimetric Analysis)

EPA Method 365.1

1.0 SCOPE AND APPLICATION

1.1 This method determines orthophosphate (PO_4^{3-}) in drinking, ground, surface, domestic waters and industrial waste.

1.2 The applicable range of this method is 0.004 to 0.250 mg P/L.

2.0 SUMMARY OF METHOD

The orthophosphate ion (PO_4^{3-}) reacts with ammonium molybdate and antimony potassium tartrate under acidic conditions to form a complex which is further reduced by ascorbic acid to form a blue complex, which absorbs light at 880 nm. The absorbance is directly proportional to the concentration of orthophosphate present in the sample.

3.0 INTERFERENCES

3.1 Silica forms a pale blue complex, which also absorbs at 880 nm. This interference is insignificant on the silica concentration of 1 mg SiO_2/L. If the silicate concentration is higher than 1 ppm, the sample result will not be reliable within the calibration range of the method.

3.2 Concentrations of ferric iron (Fe^{3+}) greater than 50 mg/L will cause a negative error due to precipitation of and subsequent loss of orthophosphate.

3.3 Method interferences may be caused by contaminants in the reagent water, reagents, glassware, and other sample processing apparatus that can bias analyte response especially in low level detection of OP. To eliminate this problem wash glassware with 1:1 HCl and rinse with DI water.

4.0 HEALTH AND SAFETY

4.1 Accepted laboratory safety practices should be followed during reagent preparation and instrument operation. The use of a fume hood, protective eyewear, lab coat and proper gloves is required when preparing reagents.

4.2 The following chemical has the potential to be highly toxic or hazardous.

4.2.1 Sulfuric Acid

4.3 A reference file of Material Safety Data Sheets (MSDS) is available to all personnel involved in the chemical analysis.

5.0 EQUIPMENT AND SUPPLIES

5.1 Equipment

5.1.1 Lachat Quick Chem FIA 8500 series.

5.1.1.1 XYZ Auto sampler ASX-520 series with sample, standard and dilution racks

5.2.1.2 Manifold or reaction unit

5.2.1.3 Multichannel Reagent Pump RP-100 series

5.2.1.4 Colorimetric Detector

5.2.1.4.1 Flowcell, 10 mm, 80uL, glass flow cell

5.2.1.4.2 880 nm interference filter

5.2.1.5 Computer, monitor, printer and The Flow Solution software.

5.2 Supplies

5.2.1 13x100 mm test tubes, Fisher # 14-961-27

5.2.2 16x125 mm test tubes, Fisher # 14-961-30

6.0 REAGENTS AND STANDARDS

6.1 Reagents

6.1.1 Stock Ammonium Molybdate Solution- In a 1 L volumetric flask, dissolve 40.0 g ammonium molybdate tetrahydrate [$(NH_4)6Mo7O_{24}.4H_2O$] in approximately 800 ml DI water. Dilute to the mark and let stir for 4 hours. Store in a plastic and refrigerate. May be stored up to two months when kept refrigerated.

6.1.2 Stock Antimony Potassium Tartrate Solution- In a 1 L volumetric flask, dissolve **3.22 g antimony potassium tartrate Trihydrate $K(SbO)C_4H_4O_6.3H_2O$)** or dissolve **3.0 g antimony potassium tartrate hemihydrate $K(SbO)C_4H_4O_6.1/2H_2O$),** in approximately 800 ml DI water. Dilute to the mark and let stir for few minutes. Store in a dark bottle and refrigerate. This stock may be used up to two months when kept refrigerated.

6.1.3 Molybdate color Reagent, 1 L- Add carefully, while mixing, 35 ml sulfuric acid to about 500 ml DI water. When the temperature is cool add 72.0 mL Stock Antimony potassium Tartrate and 213 mL Stock Ammonium Molybdate Solution. Dilute to the mark and invert three times. Degas with helium for 2 minutes. Prepare fresh weekly.

6.1.4 Ascorbic Acid Reducing Solution, 0.33 M – In a 1 L volumetric flask dissolve 60.0 g granular ascorbic acid in about 700 ml DI water. Add 1.0 g dodecy sulfate (CH_3 $(CH_2)_{11}OSO$ 3Na). Use degassed water to prepare this reagent. Prepare fresh weekly. Discard if the solution becomes yellow.

6.1.5 Sodium Hydroxide – EDTA Rinse - Dissolve 65 g sodium hydroxide (NaOH) and 6 g tetrasodium Ethylenediamine tetraacetic acid (Na4EDTA) in 1.0L DI water.

6.2 Standards

6.2.1 Orthophosphate Stock Standard (1000 mg P/L) - This standard is pre-made and purchased from RICCA CHEMICALS (cat. no. 5839.1-16). If this stock standard is not available, prepare by dissolving 4.396 g of primary standard grade anhydrous potassium phosphate monobasic (KH2PO4) that has been dried in the oven for one hour at 105 ^{o} C in about 500 ml of DI water. Bring up to 1000 mL mark with DI water and store at 4^{o} C. Prepare this reagent monthly.

6.2.2 Intermediate Standard (50 mg P/L) - Pipette 5 ml of standard 6.2.1 into a 100 ml volumetric flask. Bring up to mark with DI water. Store at 4^{o} C. Make weekly.

6.2.3 Spiking Solution (50 mg P/L) - This is the same as the intermediate standard, which is used to spike the samples. Pipette 30 uL of the spiking solution (standard 6.2.2) into 10 mL of DI water or 10 mL of sample, in order to make the blank spike and sample spike. The concentration value for spiking solution is 0.15 mg/L.

6.2.4 Working Standards - The working standards are prepared weekly according to the following table:

Orthophosphate ppm	**Combined Intermediate Std**	**Final Volume**
0.000	DI water	100 ml
0.004	2.67 ml of std 0.150 ppm	100 ml
0.010	20 mL of std 0.050 ppm	100 ml
0.050	100 uL	100 ml
0.100	200 uL	100 ml
0.150	600 uL	200 ml
0.200	400 uL	100 ml
0.250	500 uL	100 ml

7.0 SAMPLE COLLECTION, PRESERVATION, AND STORAGE

7.1 Samples are collected in plastic or glass bottles. All bottles must be thoroughly cleaned and rinsed with reagent water.

7.2 Never use acid preservation for samples to be analyzed for HL OP.

7.3 Samples to be analyzed for Orthophosphate only are cooled to $4^{o}C$ and analyzed within 48 hours. For short-term preservation freeze at $-20^{o}C$.

8.0 QUALITY CONTROL

8.1 An initial Demonstration of Performance (IDP) to establish the instrument's capability is required prior to its use in analyzing client's samples. The linearity of calibration range (LCR) and the ability to quantify the Quality Control Samples correctly are used to assess the performance.

8.2 A demonstration of capability study is performed by each analyst performing the test by analyzing four replicates of an external quality control sample during one or two analytical runs.

8.3 Immediately following daily calibration, a mid-range check standard and a calibration blank is analyzed, also after every ten samples (or more frequently, if required) and at the end of run. If a check standard fails, the samples between the last satisfactory check standard and the unsatisfactory standard are re-analyzed.

8.4 Every tenth sample is duplicated (analyzed from two different tubes) and spiked. The accepted value for the relative Percent Difference (RPD) or spike recovery is ± 10 %. If these do not fall within the accepted ranges, the corresponding analyses are repeated.

8.5 A known QC sample for Orthophosphate is run in the beginning and at the end of each run.

8.6 A method detection limit (MDL) study is performed once a year by analyzing seven replicates of the 0.004 ppm standard spread over three analytical runs. MDL is calculated as follows:

MDL = (t) x (S) where, t = Student's t value for a 99% confidence level and a standard deviation estimate with n-1 degrees of freedom [t = 3.14 for seven replicates] and S = standard deviation of the replicate analyses.
Each new analyst also performs an MDL study or when any changes in the analytical procedure or instrument are made.

8.7 All the standards are run in duplicates: that is, two analyses from each tube. A linear calibration is performed daily before the sample run.

9.0 PROCEDURE

9.1 Sample preparation

9.1.1 Prepare a list of samples to be analyzed and pour samples into labeled tubes (16mm x 125 mm test tubes).

9.1.2 Spike every tenth sample by adding 30 uL of 50 ppm P/L (Intermediate Standard) into 10 mL DI water or 10 mL of sample.

9.1.3 Filter the turbid samples by inserting the Sera Filter inside the 16 X 125 mm test tubes containing the sample. Press the filter down and pour the filtered sample collected on the top inside a 13 x 100 mm test tube for analysis.

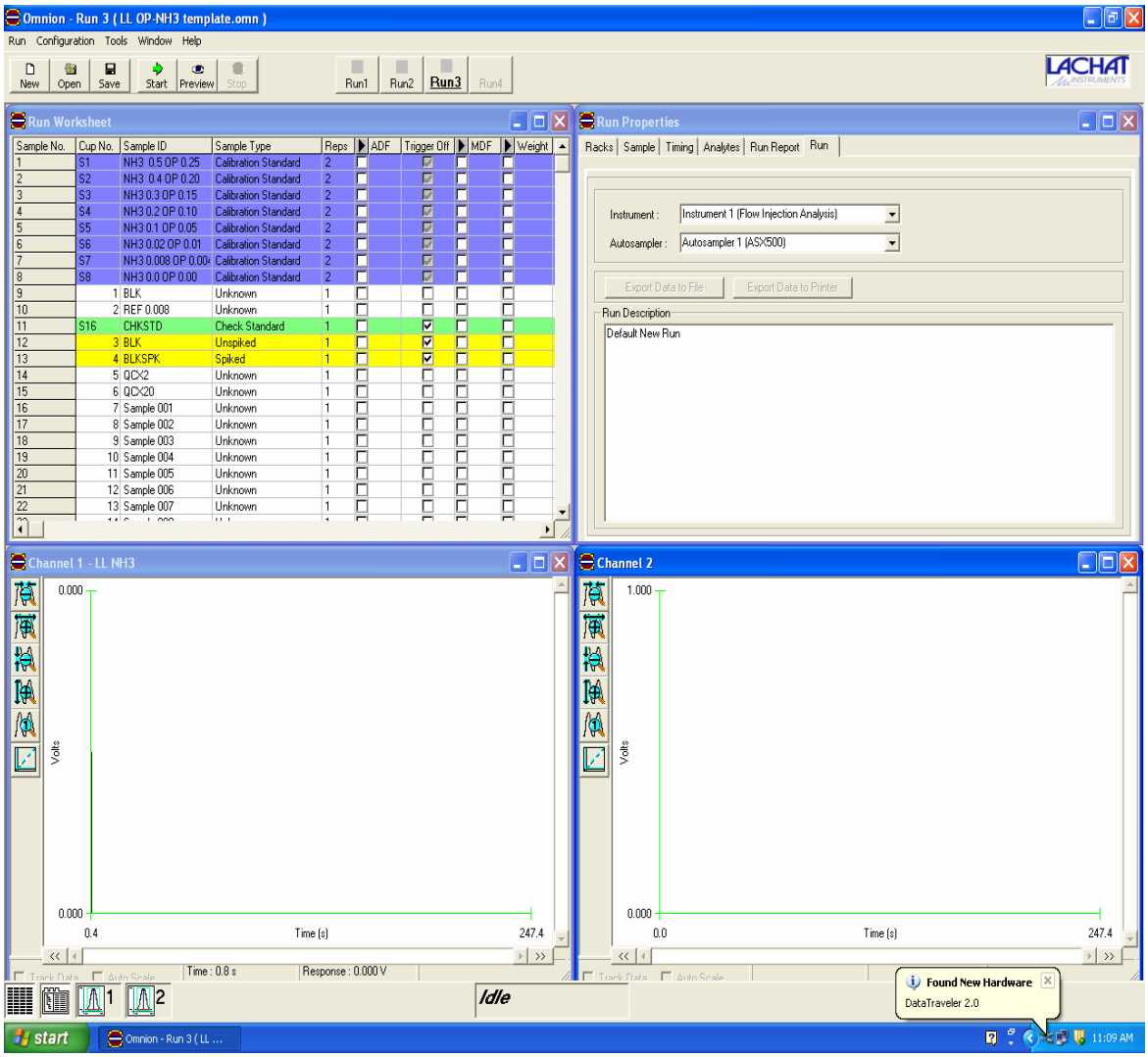
9.1.4 To prevent bubble formation, degas all reagents, except those specified by the method with helium. Use He at 140 kPa (20lb/in2) through a helium degassing tube or a pipette for two minutes.

9.2 Instrument set-up and sample analysis

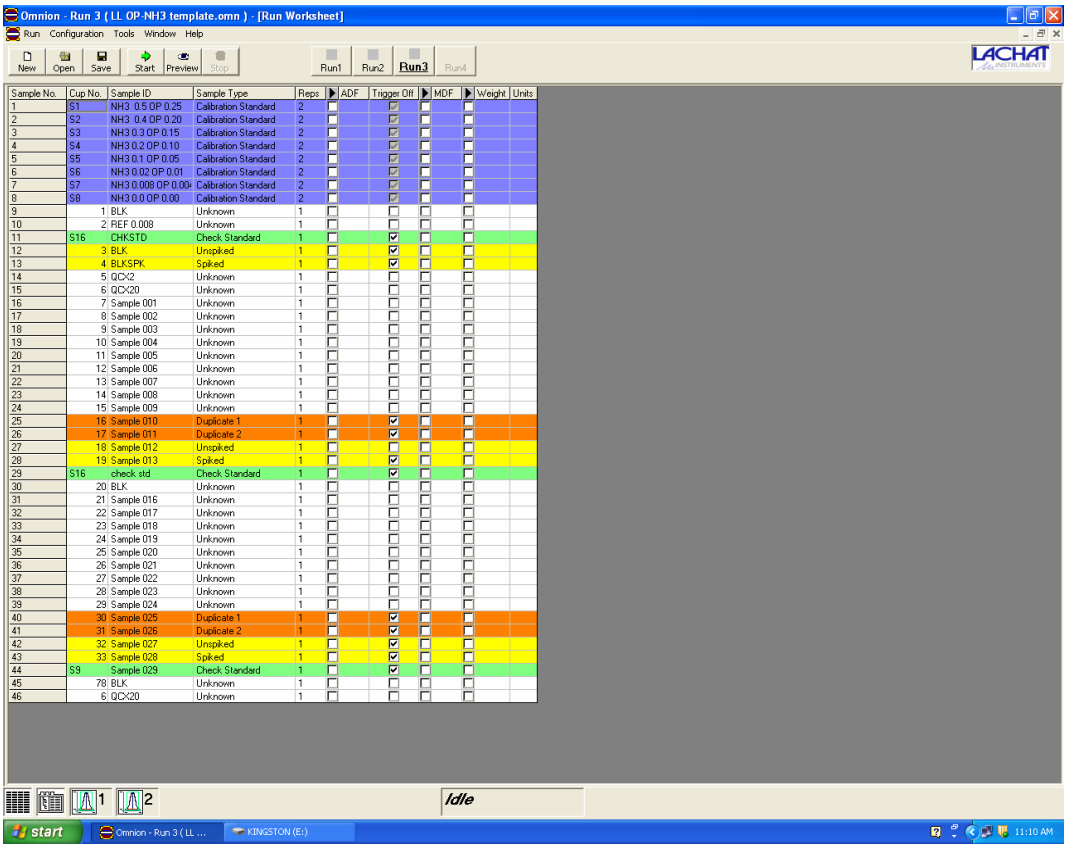
9.2.1 Set up manifold as in the diagram.

9.2.2 Turn on the Lachat instrument, computer, monitor, and printer.

9.2.3 Double click on **"LL OP/NH3"** to open the template, which consists of four windows. Samples are analyzed consecutively for orthophosphate and ammonia on the same system.



9.2.4 Maximize the **"Run Worksheet"** window at the top left hand corner of the screen by clicking on the middle square on that screen to open the worksheet template. Enter sample identifications in the sample ID column, making sure that all duplicates and spikes are entered in the appropriate locations of the worksheet. Identifications of the quality control samples analyzed at the beginning of the run and the locations of the duplicates and the spiked have been saved on the template. Press **"Enter"** key after each entry in order to save all entries.

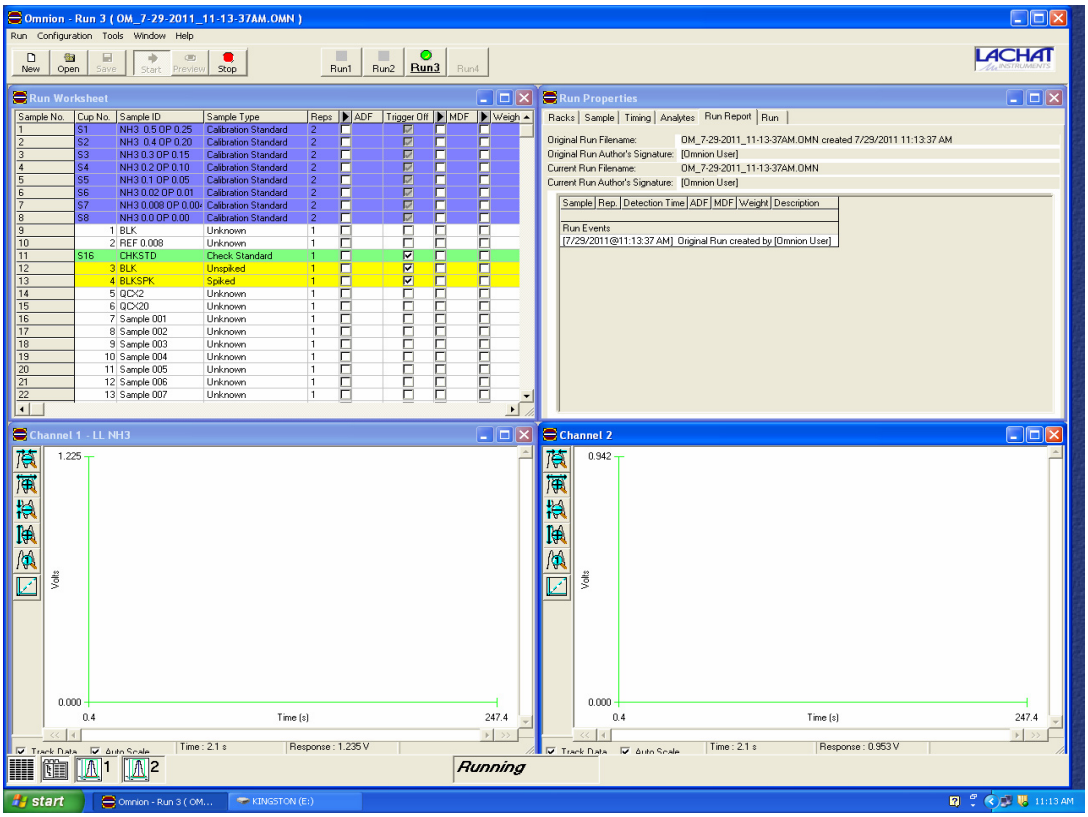


9.2.5 Print a copy of this worksheet by first double clicking on **"Run"** icon and then selecting **"Export Worksheet Data"**.

9.2.6 Click on **"Window"** tab and then, click on **"Tile"** to return to the screen with three windows.

9.2.7 Place standards in standard vials in the standard rack in order of decreasing concentration in positions 1 to 8 (STD 8 is DI water-0.00 mg/L). Place the check standard vial in position 16 (S16) of the standard rack. Pour samples and quality control samples into 13 x 100 mm test tubes and place them in the sample racks according to the worksheet prepared in 9.2.4.

9.2.8 Pump deionized water through all reagent lines for 15 – 20 minutes and check for leaks and smooth flow. Switch to reagents and continue pumping for about 10 minutes. Click on **"Preview"** tab to monitor the baseline.



9.2.9 Once a stable baseline is achieved, click on **"Stop"** tab to stop monitoring the baseline. Click on **"Start"** tab to begin the analysis.

9.2.10 If the calibration passes, instrument will continue to analyze the samples. If it fails, take appropriate corrective actions and recalibrate before proceeding to analyze samples.

9.2.11 Manual dilution will be performed to reanalyze samples with concentration exceeding the calibrated range.

9.2.12 After the run is complete, remove the reagent lines and place them in DI water and rinse for about 15 minutes. If necessary,
rinse the OP channel with the NaOH – EDTA rinse solution (6.1.5) for about 5 minutes followed by DI water for 10 – 15 minutes. Pump the tubes dry, release the pump tubing, and turn off the pump.

10.0 DATA ANALYSIS AND CALCULATIONS

10.1 All the calculations are performed by the Omnion 3.0 software system. It uses a calibration graph of peak area versus concentration to calculate the phosphorus concentrations in the samples. All standards are analyzed in duplicate and all data points are used for the calibration curve. Samples with phosphorus concentrations greater than 0.250 ppm are manually diluted and reanalyzed.

10.2 Calculate the percentage spike recovery of the laboratory fortified samples as follows:

$$\%\,SR = \frac{(\text{spiked sample conc.} - \text{sample conc.}),\ \text{ppm}}{\text{amount of spike added to sample, ppm}} \times 100$$

10.3 Calculate the relative percentage difference for the duplicated samples as follows:

$$\%\,RPD = \frac{\text{difference between the duplicates}}{\text{average of the duplicates}} \times 100$$

11.0 DATA AND RECORDS MANAGEMENT

11.1 Completed data packages are scanned and stored electronically before being placed in the appropriate binders in the lab.

11.2 Report only those results that fall between the lowest and the highest calibration standards. Samples exceeding the highest standard are manually diluted and reanalyzed.

11.3 Sample results for OP are reported in mg P/L. Report results to three decimal places. Report results below the lowest calibration standard as <0.004. For the Chesapeake Bay Program, only report all calculated results with the "L" sign for concentrations less than that of the lowest Standard.

12.0 WASTE MANAGEMENT

12.3 It is laboratory's responsibility to comply with all state, federal and local regulations governing waste management, particularly the hazardous waste identification rules and disposal restrictions, and to protect the air, water and land buy minimizing and controlling all releases from fume and bench operation.

12.4 Samples and standards are poured down the drain while large amount of water is running. For more information consult the "Waste Management manual for Laboratory Personnel", available from the American Chemical Society's Department of Government Regulations and Science Policy, 1155 Street N. W., Washington D. C. 20036, (202) 872-4477.

13.0 REFERENCES

13.1 EPA Method 365.1, *Methods for the Determination of Inorganic Substances in Environmental Samples*, August 1993.

13.2 American Public Health Association, *Standard Methods for the Examination of Water and Wastewater,* 21st Edition, Method 4500- P E, 2005.

13.3 *Lachat Instruments QuickChem Method 10 – 115 – 01 – 1 – A9,* Determination of Orthophosphate by Flow Injection Analysis.

13.4 Lachat Instruments, *Operating Manual for the Quikchem Automated Ion Analyzer*.

13.5 Division of Environmental Chemistry, DHMH-Laboratories Administration, *Quality Assurance Plan,* Revision 14.0, October 2014

13.6 Division of Environmental Chemistry, DHMH-Laboratories Administration, *Quality Manual,* Revision 1.0, November 2014

APPENDIX A

Division of Environmental Chemistry
INORGANICS ANALYTICAL LABORATORY

Data Review Checklist LL Orthophosphate

EPA Method 365.1

LabNumbers[1]:_____

Date Collected: _____ Date Analyzed: _____Analyst: _____

Procedure	Acceptance Criteria	Status (✓)	Comments
Holding Time	48 hours @ 4°C/ 28 days @ –20°C		
Calibration Curve	Corr. Coefficient. ≥ 0.9950		
Reagent Blank	< Reporting level (0.004 ppm)		
Blank Spike	1 per batch		
	Recovery = 90–110%		
Matrix Spike	Every 10th sample or 1/batch, if less than 10 samples		
	Recovery = 90–110%		
External QC	Beginning and end of each run		
	Within acceptable range		
Check Standard	After every 10th sample and at the end of the run		
	Concentration = 90–110% of the true value		
Duplicates/Replicates	Every 10th sample or 1/batch, if less than 10 samples		
	RPD ≤ 10%		
Decimal Places Reported	3		
Measured Values	Within calibration range (0.004–0.250 ppm)		
Diluted Samples	Correct final calculations		
Changes/Notes	Clearly stated		

[1]Include beginning and ending numbers, account for gaps by bracketing.

_____ _____
Analyst's Signature & Date Reviewer's Signature & Date

_____
Supervisor's Signature & Date

Reagents ID
Color Reagent _____
Ascorbic Acid _____

External QC
Identification = _____
True Value = _____ ppm
Range = _____ ppm

APPENDIX B

Division of Environmental Chemistry

INORGANICS ANALYTICAL LABORATORY

Data Review Checklist LL Orthophosphate/ LL Ammonia

EPA Method 365.1/ EPA Method 350.1

Lab Numbers: [1] _____

Analyst: _____

Dates Collected: _____ **Date Analyzed:** _____

Procedure	Acceptance Criteria	Status(✓)	Comments
Holding Time	48 hours @ 4°C; 28 days @ –20°C		
Calibration Curve	Corr. Coeff. ≥ 0.9950		
Reagent Blank	< Reporting Level (0.004 ppm for OP; 0.008 ppm for NH_3)		
Blank Spike	1 per batch		
	Recovery = 90–110%		
Matrix Spike	Every 10th sample or 1/batch, if less than 10		
	Recovery = 90–110%		
External QC	Beginning and end of each run		
	Within acceptable range		
Check Standard	After every 10th sample and at the end of the		
	Concentration = 90–110% of the true value		
Duplicates/Replicates	Every 10th sample or 1/batch, if less than 10		
	RPD ≤ 10%		
Decimal Places Reported	3		
Measured Values	Within calibration range (0.004–0.250 ppm for OP; 0.008–0.500 ppm for NH_3)		
Diluted Samples	Correct final calculations		
Changes/Notes	Clearly stated		

[1] Include beginning and ending numbers, account for gaps by bracketing.

_____ Analyst's Signature & Date

_____ Reviewer's Signature & Date

_____ Supervisor's Signature and Date

NH_3 Reagents	ID	OP Reagents	ID
Sodium Phenolate		Color Reagent	
Sodium Nitroprusside		Ascorbic Acid	
Sodium Hypochlorite			
EDTA Buffer			

External QC

Identification = _____

True Value = _____ $_{NH_3}$ / _____ $_{OP}$ ppm

NH_3 Range = _____ ppm

OP Range = _____ ppm

Appendix IV: Maryland Department of Health and Mental Hygiene: Divisional Analytical Corrective Action Form

State of Maryland
DHMH - Laboratories Administration
DIVISION OF ENVIRONMENTAL CHEMISTRY

DIVISIONAL ANALYTICAL CORRECTIVE ACTION FORM

Quality Assurance Program

❒ **NONCONFORMANCE**

Customer		**Samples(s)** E1400326800l	
Test:	**Method:**	**Instrument:**	**Date of Occurrence:**

---- Failed Tuning	---- Power Failure	---- Exceeded Holding Time
---- Failed Calibration	---- Broken or Lost Aliquot	---- Matrix Interference
---- Instrument Instability	---- Insufficient Volume	---- Out-of-Control QC Param.
---- Instrument Malfunction	---- Poor Aliquot Preservation	
-✓ Other		

Detailed Description	
Signature of Originator	**Date**

❒ **CORRECTIVE ACTION TAKEN**

---- Instrument Returned ---- Instrument Recalibrated ---- Instrument Serviced	---- Sample(s) Re-poured ---- Sample(s) Reanalyzed ---- Lab Management Notified -✓- Other

Detailed Description	
	Date of Completion:
Signature of Person Responsible	**Date**

❒ **VERIFICATION OF NONCONFORMANCE AND CORRECTIVE ACTION**

Signature of Supervisor	**Date**

❒ **NOTIFICATION**

Customer Contact Required? ---- Yes --- No	**SMA / Date of Contact**
Detailed Description	
Signature of Notifier	**Date**

❒ **IS FURTHER INVESTIGATION / MONITORING NEEDED?** ☐ **YES** ☐ **NO**

If YES, Please Forward To	Date

❒ **DESCRIBE RESULTS OF FURTHER INVESTIGATION**

❒ **WAS PROBLEM FINALLY CORRECTED?** ☐ **YES** ☐ **NO**

If It Was Not Corrected, Explain

Corrective Action Reviewed By Supervisor	Date

Corrective Action Reviewed By Division Chief	Date

Corrective Action Reviewed By QA Officer	Date

❒ **ACKNOWLEDGEMENT**

Signature of QA Officer	**Date**

Copies: QA Officer
Laboratory QA File