Maryland Department of Natural Resources
Non-tidal Network Program
Nutrient and Sediment Load Trend Monitoring
Quality Assurance Project Plan
July 1, 2013 – June 30, 2014
Section 117(d)

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Preface

Maryland's Non-tidal Water Quality Monitoring Network began in July 2005 with funding from EPA Section 117(d). The network, presently, is composed of 21 sites where nutrient and sediment concentrations are sampled on a fixed monthly basis and eight times per year during high flow events. All sampling sites are located near a USGS stream gage, have a nearby bridge from which high flow samples may be obtained, are located at the outlet of major basins and represent watersheds with relatively high loads of nutrients and sediments.

Maryland's network was created as part of a coordinated effort, conducted by the Chesapeake Bay Program's Non-tidal Workgroup, to include all of the Chesapeake Bay watershed states in a network of stations with comparable collection and analysis protocols. Data from the Non-tidal Network will be used to estimate nutrient and sediment loads and trends in concentration for watershed management assessment purposes and for input to the CBP Watershed Model.

It is anticipated that the Non-tidal Network may be expanded spatially and upgraded as more funding becomes available.

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ACRONYMS AND ABBREVIATIONS

AMQAW - Analytical Methods and Quality Assurance Workgroup (a workgroup of the Chesapeake Bay Program's Monitoring Subcommittee)
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
DHMH - Maryland Department of Health and Mental Hygiene
DNR - 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

Distribution List

Mary Ellen Ley, USGS/Chesapeake Bay Program
Peter Tango, USGS/Chesapeake Bay Program
Rich Batiuk, EPA/Chesapeake Bay Program
Bruce Michael, MDNR
Sally Bowen, MDNR
Anthony Prochaska, MDNR
T. Mark Trice, MDNR
William D. Romano, MDNR
Scott Phillips, USGS

PROJECT MANAGEMENT

A4 Project/Task Organization

This section lists the individuals and organizations responsible for the major aspects of the Maryland Non-tidal Network Program. The flow of project tasks is indicated in Figure 1.

Director and Principal Investigator: Bruce Michael, Resource Assessment Service, DNR. 410-260-8627, bmichael@dnr.state.md.us

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, implementation, and data analysis of the program.

Quality Assurance Officer: Bruce Michael, Resource Assessment Service, DNR. 410-260-8627, bmichael@dnr.state.md.us

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

Field Sampling Operations: Sally Bowen, Project Chief, Monitoring Field Office. Monitoring and Non-tidal Assessment, DNR. 410-990-4528, sbowen@dnr.state.md.us

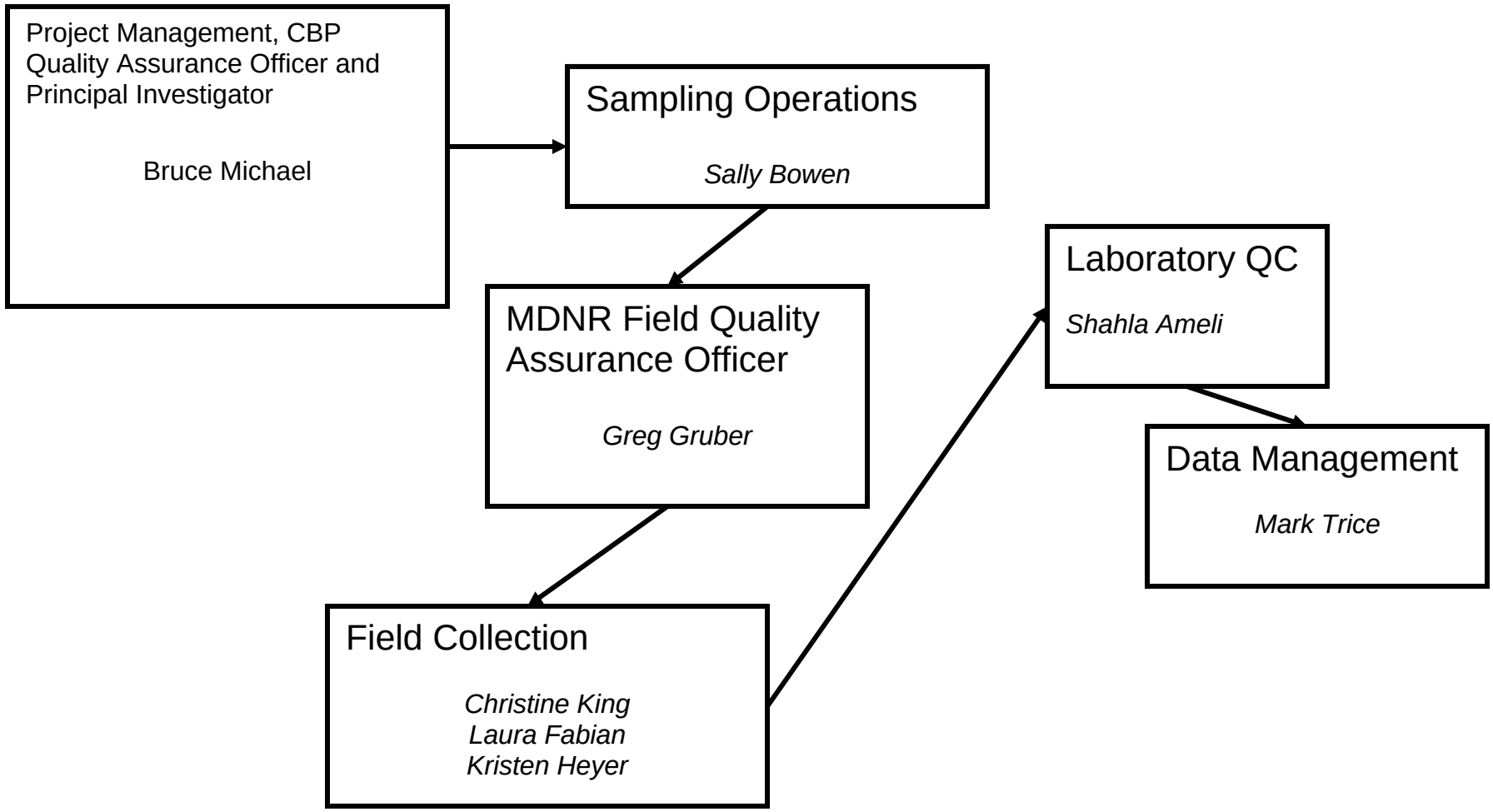
Responsibilities: This individual is responsible for administration of the field sampling activities including sample collection, sample storage, and sample delivery to laboratories.

Laboratory Analyses/Water Column Chemistry: Shahla Ameli, Inorganics Analytical Laboratory Supervisor, DHMH. 410-767-6190, amelis@dhmh.state.md.us.

Responsibilities: This person oversees the laboratory that does all of the nutrient analysis and water chemistry for the project.

Data Management; Mark Trice, Maryland Department of Natural Resources. 410-260-8649, MTrice@dnr.state.md.us

Figure 1. Organization Chart for the Maryland 117(d) Non-tidal Network Project



Responsibilities: This individual 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.

Sediment Analysis: Aimee Downs. USGS Kentucky Water Science Center. 502-493-1944, acdowns@usgs.gov .

Responsibilities: Determine suspended sediment concentration and sand/fine fractions on high flow event samples.

A5 Project Definition/Background

The Chesapeake Bay Program, under the Chesapeake 2000 agreement, is committed to reduce nutrient and sediment inputs into Chesapeake Bay. Nutrient and sediment allocations have been developed for tributary basins within the Bay watershed. This project is part of the cooperative effort of the Chesapeake Bay Program Non-tidal Workgroup to provide comparable data to assess progress in meeting nutrient and sediment reduction goals to meet water quality criteria in the Chesapeake Bay.

The main objectives of this monitoring program are to improve measurement of nutrients and sediment concentrations for the calculation of loads discharged to the Chesapeake Bay watershed, improve the accuracy of the watershed model, and help identify factors affecting nutrient and sediment loads. The project requires all participants to collect surface water samples by a method that generates horizontally and vertically integrated composite samples. This standard USGS collection method should provide data that better represents the concentration of nutrients and sediments. Since this project will specifically collect samples from eight storm events per year per station, our estimates of sediment loads contributed by Maryland's ten tributary strategy basins should be greatly improved.

Historically, Maryland's collection method at non-tidal sites was a single point grab sample. We will continue to collect a single point grab sample under our CORE/TREND network at historically collected locations. CORE/TREND sampling is conducted at co-located stations on different dates than the non-tidal network sites. We will collect an integrated sample every month under the Non-tidal Network using the USGS isokinetic sampling device if maximum velocity is 1.5 ft/sec or higher. A review of USGS discharge records indicates that for the majority of our stations, base and maximum flow is normally under 1.5 fps. Under low velocity conditions, a horizontally and vertically integrated composite sample will be generated. We anticipate that we will only collect isokinetic, equal-width increment samples during significant storm events. The stations where maximum velocity sometimes exceeds 1.5 fps during base flow are ANT0047, WIL0013, GEO0009 and, perhaps, DER0015.

Monthly measurements of in-situ parameters have also indicated that all sites except WIL0013 are well mixed. Since most of our sites are less than 110 feet wide, are 3 feet or less in depth and have a velocity under 1.5 fps, our historical data set may be a reasonable representation of dry weather conditions. When using the historical data, discarding dates with an elevated discharge reading would provide data that is more representative of baseflow conditions.

Monthly sampling is performed at all sites. Sites classified as “primary” have, in addition to monthly sampling, approximately 8 samples taken per year during high flow events (defined in this project as at least a two-fold increase in flow). These additional “storm” samples provide site-specific information on the relationship between discharge and concentration necessary for load estimations. Site selection for both primary and supplemental (monthly sampling only) was based upon 1) the presence of an operating stream gage, 2) sites located at the outlet of a tributary strategy basin, 3) sites representing high load watersheds, 4) watershed area, and 5) the presence of a bridge for sampling during high flow events.

A6 Project Task/Description

Sample collection and analysis began in July 2005 and is expected to continue for at least five years provided there is continued funding from federal and state agencies. The network currently contains 21 primary (load) sites (Table 1). Routine (pre-determined date) monthly sampling is conducted at all sites. Eight samples collected during high flow conditions will be taken at all sites each year, preferably two storms per season. Storm events are targeted because large amounts of nutrients and sediments are moved during high flows. Routine sampling is conducted over a wide range in flow conditions to provide unbiased load calculations. Sites in the Maryland CBP Non-tidal network are shown on Figure 2.

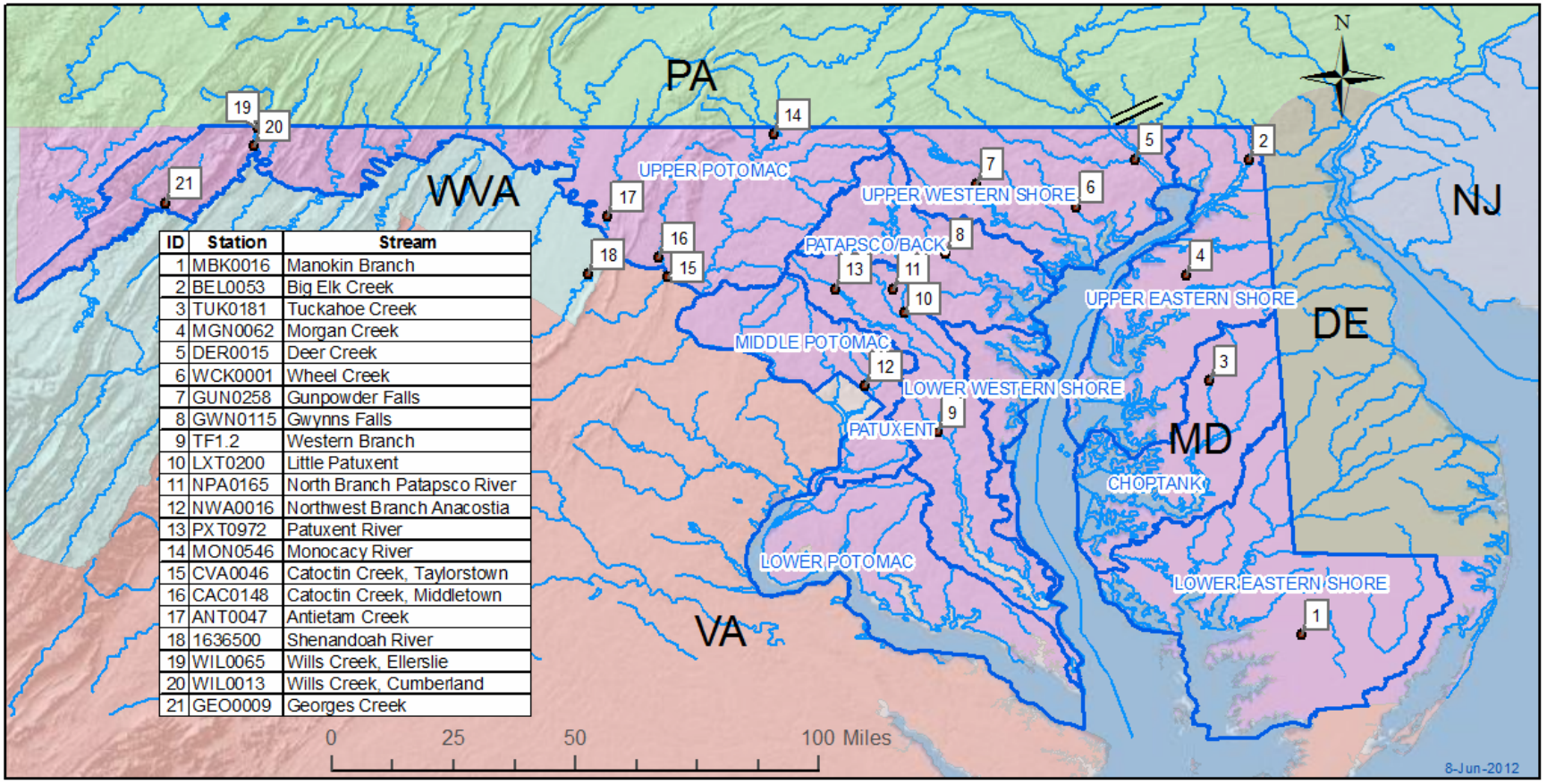
Depth integrated samples will be taken at equal width intervals and composited at all sites any time the site is sampled for the non-tidal network. Parameters and analytical methods are listed in Table 2. An additional sample will be taken from a churn splitter for sediment concentration any time high flow conditions are sampled. A sand/fine analysis will be performed on the sediment sample on a quarterly basis.

The data collected under this project will be stored on a Maryland DNR server and submitted annually to the CBP for inclusion in the non-tidal network water-quality database of the Chesapeake Information Management System (CIMS). The data are also provided to staff of the U.S. Geological Survey (USGS). USGS staff will combine the concentration data collected by DNR with stream flow data to calculate nutrient and sediment loads using the USGS Estimator program. USGS will calculate loads on an annual basis after enough years of data have been collected to accurately estimate the

model parameters predicted by Estimator. The loads can be used in CBP indicators and to demonstrate any improvements or deterioration in water quality to the public for the non-tidal network sites.

TABLE 1. Maryland CBP Non-tidal Network Program

Map #	MDNR Station ID	Stream Name	Lat (NAD83)	Long (NAD83)	Description of Sampling Location	USGS Gauge #	Network Station Type
1	GEO0009	Georges Creek	39 29.6183083	079 02.6819417	Victory Street in Westernport, MD	01599000	Primary
2	WIL0013	Wills Creek	39 39.7110433	078 46.8174567	Locust Grove Road crossing Wills Creek in Cumberland, MD	01601500	Primary
3	ANT0047	Antietam Creek	39 27.240000	077 43.965	Burnside Bridge Road near Sharpsburg	01619500	Primary
4	CAC0148	Catoctin Creek	39 19.9069327	077 34.8107379	At bridge on MD 17	01637500	Primary
5	MON0546	Monocacy River	39 41.7870000	077 14.368000	Bullfrog Road crossing the Monocacy	01639000	Primary
6	PXT0972	Patuxent River	39 14.3584867	077 03.3713467	At bridge on MD 97 near Unity	01591000	Primary
7	NPA0165	North Branch Patapsco River	39 28.9671333	076 52.9250800	Upstream of bridge at MD 91	01586000	Primary
8	GWN0115	Gwynns Falls	39 20.5671783	076 43.5833000	At bridge on Essex Road in Villa Nova	01589300	Primary
9	GUN0258	Gunpowder Falls	39 33.0386351	076 38.1520258	Confluence of Upper and Lower Glencoe Road at girder bridge	01582500	Primary
10	DER0015	Deer Creek	39 37.4085651	076 09.8863317	Bridge at Stafford Bridge Road	01580000	Primary
11	TUK0181	Tuckahoe Creek	38 58.0280000	075 56.5870000	Tuckahoe Creek at Crouse Mill Rd.	01491500	Primary
12	TF1.2	Western Branch	38 48.8580017	076 45.05207	At bridge on Water St. in Upper Marlboro	01594526	Primary
13	BEL0053	Big Elk Creek	39 37.2870000	075 49.7160000	Big Elk Creek at Rt. 279	01495000	Primary
14	MKB0016	Manokin branch	38 12.833333	75 40.283333	Near Princess Anne, MD	01486000	Primary
15	MGN0062	Morgan Creek	39 16.801667	76 0.873333	Near Kennedyville, MD	01493500	Primary
16	WCK0001	Wheel Creek	39 28.903333	76 20.431667	Near Abingdon, MD	0158175320	Primary
17	NWA0016	NW Branch Anacostia River	38 57.14	76 57.963333	Near Hyattsville, MD	01651000	Primary
18	LXT0200	Little Patuxent	39 10.065	76 51.075	Near Guilford, MD	01593500	Primary
19	WIL0065	Wills Creek	39 43.11	78 46.258333	Near Ellerslie, MD	01601100	Primary
20	CVA0046	Catoctin Creek	39 15.292	77 34.606	At Taylorstown, VA	01638480	Primary
21	NA	Shenandoah River	39 16.916667	77 47.66667	Near Millville, WV	01636500	Primary



ID	Station	Stream
1	MBK0016	Manokin Branch
2	BEL0053	Big Elk Creek
3	TUK0181	Tuckahoe Creek
4	MGN0062	Morgan Creek
5	DER0015	Deer Creek
6	WCK0001	Wheel Creek
7	GUN0258	Gunpowder Falls
8	GWN0115	Gwynns Falls
9	TF1.2	Western Branch
10	LXT0200	Little Patuxent
11	NPA0165	North Branch Patapsco River
12	NWA0016	Northwest Branch Anacostia
13	PXT0972	Patuxent River
14	MON0546	Monocacy River
15	CVA0046	Catoctin Creek, Taylorstown
16	CAC0148	Catoctin Creek, Middletown
17	ANT0047	Antietam Creek
18	1636500	Shenandoah River
19	WIL0065	Wills Creek, Ellerslie
20	WIL0013	Wills Creek, Cumberland
21	GEO0009	Georges Creek

Figure 2. Maryland DNR non-tidal network nutrient and sediment load sites and tributary strategy basins.

TABLE 2. Parameters and Analytical Methods for the Chesapeake Bay Non-tidal Water-Quality Network

Parameters/Units	Holding Time and Condition	Method/ Reference	Method Detection Limit
Field (In situ)			
Temperature, ^{O}C	< 5 min.	Standard Methods 2550 B (2000)	0.1 ^{O}C
Dissolved Oxygen, mg/L	< 5 min.	Clark cell Standard Methods 4500-OG-2001 Hach LDO Method 10360	0.2 mg/L
pH	< 5 min.	Standard Methods 4500-H^{+} (2000)	0.1 units
Specific Conductance, umhos/cm	< 5 min.	Standard Methods 2510 (1997)	1 umhos/cm
Laboratory			
Total Nitrogen, mg/L as N	Calculated	PN + TDN	N/A
Ammonium, mg/L as N	Frozen, 28 days/ 4 ^{O}C, 48 hrs.	EPA Method 350.1*****	0.0016 mg/L
Nitrite, mg/L as N	Frozen, 28 days/ 4 ^{O}C, 48 hrs.	EPA Method 353.2*****	0.0007 mg/L
Nitrate + Nitrite, mg/L as N	Frozen, 28 days/ 4 ^{O}C, 48 hrs.	EPA Method 353.2*****	0.003 mg/L
Total Phosphorus, mg/L as P	Calculated	PP+TDP	N/A
Orthophosphate, mg/L as P	Frozen, 28 days/ 4 ^{O}C, 48 hrs.	EPA Method 365.1*****	0.002 mg/L
Total Suspended Solids, mg/L	4 ^{o}C 7 days	Standard Methods 2540D	0.8 mg/L
Suspended Sediment (storms)	Dark Room 120 days	ASTM D3977C	0.5 mg/L
Sand/Fine Particles (storms)	Dark Room 120 days	DE 35997D	0.5 mg/L
Dissolved Organic Carbon, mg/L as C	Frozen, 28 days/ 4 ^{O}C, 48 hrs.	Standard Methods 5310B	0.14 mg/L
Total Dissolved Phosphorus, mg/L as P	Frozen, 28 days/ 4 ^{O}C, 48 hrs.	alk. Persulfate then EPA Method 365.1	0.006 mg/L
Total Dissolved Nitrogen, mg/L as N	Frozen, 28 days/ 4 ^{O}C, 48 hrs.	Standard Methods 4500-N C	0.034 mg/L
Particulate Carbon, mg/L as C	Frozen 28 days	Exeter Analytical Model CE-440 Elemental Analyzer	0.2233 mg/L
Particulate Nitrogen, mg/L as N	Frozen 28 days	Exeter Analytical Model CE-440 Elemental Analyzer	0.0566 mg/L
Particulate Phosphorus, mg/L as P	Frozen 28 days	Filtered particulates HCl extraction, then EPA Method 365.1	0.0003 mg/L

* Dissolved parameters are prepared by filtration through a 0.7 micron glass fiber filter.

A7 Quality Objectives and Criteria

The non-tidal network is designed to provide laboratory and field data that will help the state characterize water quality in headwater areas that affect the Chesapeake Bay. Assessment of the quality of the data collected through the program can be expressed in terms of representativeness, completeness, comparability, accuracy, and precision.

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 calls for monthly sampling on a pre-determined date, which is adequate for capturing long-term annual trends in concentration. Eight additional samples taken during high flow events are intended to determine the relationship between stream discharge and the parameters of interest so that annual loads may be calculated for primary sites.

Depth integrated samples using an isokinetic sampler or a weighted bottle sampler will be used at equal width intervals, the number of which is determined from Table 3. The type of sampler used is dependent upon flow velocity with the isokinetic sampler being used if the centroid flow velocity $\geq$ 1.5 ft/sec. Depth integrated samples are composited (i.e. combined for each width interval in a 4-liter churn splitter). Sample bottles for nutrient and sediment analysis are filled from the churn splitter.

Table 3. Minimum Number of Vertical Samples at Primary Stations
(Routine and storm event samples)

Width of Waterway (ft)	Minimum # of Verticals
0-25	1
25-100	3
100-250	5
250-500	7
>500	9

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 non-tidal network program must also be of comparable quality to the data generated by other states and laboratories participating in the program. Comparability among data sets is assured through the use of consistent field methods and protocols, participation in the Analytical Methods and Quality Assurance Workgroup (AMQAW), 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 the 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.

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 support Chesapeake Bay Program management decisions, Maryland's non-tidal network monitoring program strives to provide monitoring data of known and consistent quality to the CBPO 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 for field and lab measurements of <20 percent of the coefficient of variation; accuracy goals within 80 to 120 percent, and the completeness goals of 90 percent.

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. 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 the Department of Health and Mental Hygiene's (DHMH) participation in the Chesapeake Bay Coordinated Split Sample Program (CSSP), a split sampling program in which five 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. DHMH 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 labs. When a difference occurs, discussion begins regarding techniques and potential methods changes to resolve discrepancies and identify potential problems.

Additionally, DHMH will participate two times per year in the United States Geological Survey (USGS) Standard Reference Sample Program and will permit USGS to release the results to the Chesapeake Bay Program Quality Assurance Officer. Laboratory accuracy is 90-110% recovery.

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 analyzes and provides results on the corresponding request forms for the field duplicates. DHMH performs precision calculations for laboratory duplicates, but not for the field 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 duplicates. 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.

A8 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 Standard Operating Procedures (Appendix A). Training for field personnel who collect samples for the non-tidal network monitoring program was provided in a two-day USGS/CBP sponsored workshop on techniques required for obtaining a representative sample of nutrient and suspended sediment concentration in Harpers Ferry, WV in September 2004.

Training of personnel at DHMH is conducted in the laboratory. Each new analyst is trained on the laboratory procedures he or she will be assigned to perform by an experienced analyst. Training is documented using the Division of Environmental Chemistry's training forms and signed by the Supervisor and the Division Chief.

A9 Documentation and Records

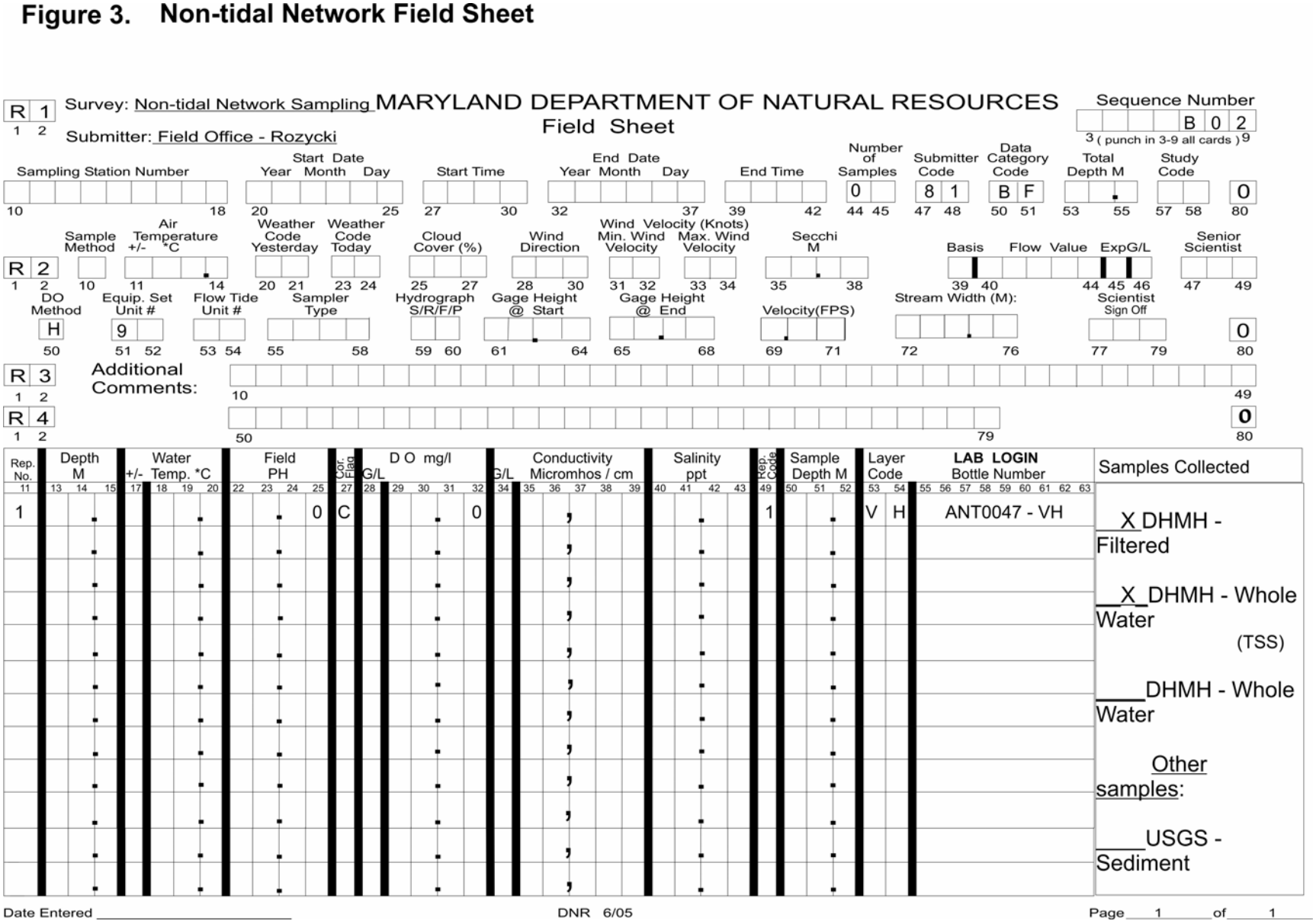
Documenting sampling events is an important component of the non-tidal network monitoring program. Field crews document all data obtained in the field on field sheets (Figure 3). Since the data generated by this program are not used for legal purposes, a formal chain of custody sheet is not required. Field sheets and any information

concerned with specific problems and/or events during a sampling run, as well as modifications to the sampling program will be maintained by field office staff.

The water quality monitoring field sheet is completed on arrival at each station. This form is used to record sample collection depth, weather, flow, field measurements (pH, water temperature, dissolved oxygen, and conductivity) and data pertinent to the collection of samples such as type of sample, and date and time of collection. A field notebook 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 (c.f. distribution list). The QAPP will be updated annually. Any modifications to the QAPP will be reviewed and approved by the personnel conducting the sampling, the principal investigators and DNR's Quality Assurance Officer. Once approved, the final version will be made available to all interested parties by placing downloadable copies in the Chesapeake Bay Program's and DNR's websites. Project reporting to management will be accomplished by semi-annual progress reports of activities. Electronic summaries of provisional instantaneous water quality data will be provided on an annual basis.

Figure 3. Non-tidal Network Field Sheet



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DATA GENERATION AND ACQUISITION

B1 Sampling Process Design

Sampling locations are described in Table 1 and shown on Figure 2. In order for a site to be considered for the non-tidal network program, it needed to: 1) have an operating stream gage, 2) represent tributary strategy basins, 3) represent watersheds with high nutrient and sediment loads, 4) be among Maryland's larger watersheds, and 5) have a safe bridge from which storm samples could be taken. Stations from DNR's historical network were reviewed to see if they met these criteria and stations in under-sampled regions of the state were also considered. Data are available for the ten initial primary sites (ANT0047, BEL0053, GWN0115, DER0015, NPA0165, TF1.2, MON0546, TUK0181, WIL0013, and GEO0009) starting July 2005. Three supplemental sites were added in October 2005 (GUN0258, PXT0972, and CAC0148). The three supplemental sites all have the same attributes as primary sites and were upgraded to primary sites with storm event sampling in January 2007. Five new primary sites (MKB0016, MGN0062, WCK0001, NWA0016, LXT0200) were added in October 2011. Three additional sites (CVA0046, WIL0065, and 01636500) were added to the non-tidal network in the spring of 2012 following receipt of Chesapeake and Atlantic Coastal Bays Trust Fund money. Given the nature of this project, the only matrix sampled at these stations is water. The parameters of interest are presented in Table 2.

Data were collected at the ten initial primary sites starting in January 2005; however, data from January 2005 through June 2005 are not included in the data base due to concerns that a step trend could be introduced following a laboratory methods change that occurred in July 2005.

Routine monitoring (pre-determined date) is conducted on a monthly basis to obtain data over a range of flows. Eight storm event samples are collected per year at each station because the highest nutrient and sediment loads are generated during high flows. Ideally, two storm event samples are collected in each season. This sampling protocol was designed to provide adequate data for use in the U.S. Geological Survey Estimator model, which is used to calculate loads and trends.

B2 Sampling Methods

Sampling protocols for the Maryland non-tidal network can be found in the Maryland *Non-Tidal Network Program Standard Operating Procedures* (Department of Natural Resources, 2009) (Appendix A). Routine and storm event samples are collected using a modified version of the U.S. Geological Survey equal width interval method that reduces the number of vertical samples from ten to an odd number based on the width of the stream being sampled (Table 3).

Routine samples: Equal width interval, depth integrated samples are taken monthly at

primary sites. Vertical sampling is done with a DH-81 if the stream is wadable or a DH-95 (a DH-59 was used in 2005) where samples are accessible only from a bridge. A weighted bottle, WBH-96, is used when the velocity is less than 1.5 ft/second. Samples are composited in a 4-liter churn splitter from which subsamples are drawn. A single whole water sample bottle is drawn and sent to DHMH for TSS analysis. A second whole water sample bottle is drawn and field processed for dissolved nutrients and particulate analysis. A sample is also collected for total suspended sediment analysis and sent to the USGS Sediment Laboratory in Kentucky if routine sample collection occurs during a high flow event.

Storm samples: Sampling during high flow events is accomplished from a bridge by use of a DH-95 at all 21 primary sites. All vertically integrated samples are collected in the churn splitter and, in addition to the TSS, dissolved nutrients and particulate filters for analysis at DHMH, a whole water sample is drawn to send to the USGS Sediment Laboratory in Kentucky for suspended sediment analysis.

Field Measurements: Dissolved oxygen, pH, specific conductance and temperature are measured monthly and during high flow events by using a Hydrolab or YSI. Measurements are taken at the center of each equal width interval and the median value is recorded for the sample. Average stream flow during the sampling period, as reported by USGS, is also recorded. See the attached Non-tidal Network Monitoring Program Standard Operating Procedures for more detailed information.

B3 Sample Handling and Custody

With the exception of sediment samples, laboratory samples are placed on ice and transported in coolers from monitoring stations to the field office in Annapolis by Maryland Department of Natural Resources (DNR) Monitoring Program field personnel. Samples are either frozen for later delivery or taken directly to the DHMH laboratory by field personnel or left with a courier for delivery to the DHMH laboratory. Data sheets accompany these samples to DHMH (Figure 4). Sample bottles for sediment analysis are shipped to the USGS Water Science Center Sediment Laboratory in Louisville, KY on an intermittent basis. These data are not used for legal purposes, therefore chain of custody forms are not used.

Note that DNR does not currently acidify samples for ammonium (NH4), nitrate+nitrite (NO23), or dissolved organic carbon (DOC) as described in the Code of Federal Regulations (40 CFR Part 136). DNR has completed an assessment of the effects of preservation method on NH4 concentration by analyzing samples that have been preserved with sulfuric acid (acidification), freezing, or keeping the samples on ice and delivering them to the laboratory for analysis within 24 hours. The results indicated that acid-preserved field samples are biased low compared frozen samples and samples kept on ice for analysis within 24 hours. Acid-preserved samples were also biased low compared to known high and known low NH4 concentrations of samples prepared in the

laboratory. The results of this study have been documented in a data analysis issues tracking system (DAITS) report (DAITS050). DAITS050 is available on the Chesapeake Bay Program web site (http://www.chesapeakebay.net/). DAITS050 has been approved by the Analytical Methods and Quality Assurance Workgroup, therefore DNR will continue to preserve NH4 samples by keeping them on ice if they can be delivered to the lab for analysis within 24-hours, or by freezing for longer holding times. The Chesapeake Bay Program has requested written approval from EPA Region III to grant DNR an exemption from using acidification to preserve NH4 samples and is awaiting their reply.

DNR has also reviewed data that compared the effect of preservation method on NO23 and DOC. The results of that study are documented in DAITS051, which has not yet been approved by the Analytical Methods and Quality Assurance Workgroup. A statistically significant difference was detected between iced (the current method) and acid-preserved samples; however, there was far more variability in NO23 concentrations among seasons and stations than among preservation methods. Statistically significant differences were also detected among preservation methods for DOC; however a test of multiple comparisons grouped acid and iced together. Mean differences between acid and iced DOC samples were less than the method detection limit and differences between acid and frozen only slightly exceeded the method detection limit. Given the equivalency between chilling for 24 hours or freezing for longer periods and acid preservation, DNR will continue to use the historic methods, pending approval of DAITS051.

B4 Analytical Methods

All Hydrolab and YSI instruments are calibrated both prior to and after their use for measuring temperature, pH, dissolved oxygen, and conductivity. All calibration checks are recorded in field logbooks. Laboratory personnel follow EPA guidelines on quality control and quality assurance. Minimum detection limits for field measurements are described in Table 2.

R 5 study code: 0 9 Replicate:

Maryland Department of Natural Resources

Sequence Number

6 (punch in 6-13 all cards) 13

Survey: NTN: Antietam Creek

Collector: _____

AFO-410-990-4526/C. Rozycki

Laboratory Analysis Sheet (Non-tidal Network Program)

Data Code 1B

Sample Station Number: A N T 0 0 4 7

Date (Year / Month / Day) | Start Depth M: 0.0 | End Depth M | Start Time | Submitter Code: 8 0

Bottle Numbers:

Type of Sample: Whole (1 quart bottle): A N T 0 0 4 7 V H | Filtered (8 ounce bottle): A N T 0 0 4 7 V H | PC/PN/PP Foils: S E P S H E E T 0

R 6 | Sample Method: 6 | Data Category Code: B F | Sample Layer: V H | 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									
	BOD 5 day mg/l	B O D 5									
	Turbidity NTU (W)	T U R B									
	Total Alkalinity (W) mg/l	T A L K									
X	Total Susp. Solids (W) mg/l	T S S									
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 | 0

Date Entered: _____

DNR 5/2005

Send Results To:

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

Figure 4. Non-tidal Network Laboratory Analysis Sheet

B5 Quality Control

The data collected as part of the Non-tidal Network are used in making management decisions regarding Chesapeake Bay water quality as described in the Introduction. 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. General discussions of quality assurance and quality control aspects associated with accuracy, precision, and audits are provided in the subsections below.

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 five laboratories involved in Chesapeake Bay monitoring analyze the coordinated split samples.

Precision: Precision of the chemical analytical methods is determined and documented from duplicate analyses. 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.

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.

Reporting: Quality assurance information for field duplicate samples in the mainstem and tributaries is stored on the routine computerized water quality data sets as replicate observations that can be used to assess precision. Laboratory quality assurance/control information on duplicates and spikes is stored on a computerized data set at the laboratory.

B6 Instrument/Equipment Testing, Inspection, and Maintenance

Field crews carry two calibrated Hydrolab or YSI meters 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 under special remarks.

The specific meter to be used each day receives a dissolved oxygen validation check. The meter is set up for a dissolved oxygen calibration, but the oxygen value is only adjusted if drift is greater than 0.4 mg/L.

B7 Instrument/Equipment Calibration and Frequency

The procedures outlined here refer to the Hydrolab instruments. The detailed calibration procedures will be performed as described in the Hydrolab 4000, Surveyor II and Scout II Operation Instructions Manual.

I. Calibration

A. Set up a calibration logbook for each unit, with make, model, and serial number and purchase date. Assign a letter for DNR 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 any parameter has drifted.

C. Specific conductance calibration shall be made using standards generated by the field office from dry KCl and deionized water. The standard used for sampling the non-tidal waterways of Maryland is 294 microsiemens/cm (microsiemens=microS=μS); or 0.002, molar KCl (At 25 $^{\circ}$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. Dissolved Oxygen calibration shall be done one of two ways. The Hydrolab instruments outfitted with a Clark cell are calibrated 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. Hydrolab and YSI instruments outfitted with a luminescent-based dissolved oxygen probe, LDO and ROX, respectively, are calibrated using an air-saturated water method. After correcting for barometric pressure and temperature, the oxygen content of the air-saturated water can be checked against standard DO tables. The membrane (YSI) or cap (LDO) is also visually checked every time the meter is pre- or post-calibrated. If any damage is observed, the instrument is pulled for servicing and a replacement instrument is used for sampling (this is not common).

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

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

DNR does not follow the DO probe calibration procedures that are described in Chapter V (Non-tidal water quality monitoring) of the Recommended Guidelines for Sampling and Analysis in the Chesapeake Bay Monitoring Program: (http://www.chesapeakebay.net/committee_analyticalmethodsworkgroup_projects.aspx?menuitem=16701). The DO field measurement protocol calls for a calibration at the beginning and end of each day to ensure there has been no drift in the sensor. DNR conducted a year long analysis of optical DO sensor post-calibration drift data and found that generally there were differences of $\leq$0.1 mg/L DO between pre- and post-calibration readings even after a week. In 2011 DNR received approval from the Chesapeake Bay Program Non-tidal Workgroup to continue with weekly calibrations.

B8 Inspection/Acceptance of Supplies and Consumables

The deionized water at the Field Office 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 in line 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.

Deionized water blanks are submitted to the laboratory with every sampling run. The field staff rinses a 16 ounce bottle three times with deionized water directly from the storage container in the field vehicle. The blank is filled to the bottle shoulder to leave space for expansion if the samples are frozen. The blank is then submitted to the laboratory along with field samples for analysis.

DHMH uses a water system provided and serviced by SIEMENS. 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. All supplies and consumables for laboratory analyses are described in the applicable standard operating procedure section for the parameter in question (see Appendix D).

B9 Non-direct Measurements

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

B10 Data Management

Data collection for the chemical and physical properties component of the non-tidal network program begins when measurements from field recording instruments are entered onto field data sheets. A field log book is used to document any problems encountered in the field that might affect the field parameters or the samples brought back for the laboratory. A senior field scientist ensures that all measurements are taken 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 DNR 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, laboratory, and suspended sediment results to expect. See Appendix B for a sample Cross Reference Sheet and associated documentation.

Nutrient laboratory data sheets are also initiated in the field. These nutrient laboratory sheets list each parameter requested for analysis and include basic information about the sample, such as station, date, time, depth, and volume filtered. The sheets serve as sample transfer sheets, traveling with the samples to the Maryland Department of Health and Mental Hygiene laboratory (DHMH) for nutrient analysis or to the USGS

Kentucky Water Science Center for sediment analysis. Both the sheets and the samples are logged in at their respective laboratories.

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 nutrient lab 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 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. 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.

Sediment data analyzed by the USGS Kentucky lab are transmitted to DNR directly as text files. Because of the time lag between processing field and DHMH laboratory data and sediment samples, the sediment results are reviewed by a DNR biologist using a separate computer program at a later time. The sediment data management program reads data from the USGS text file and merges it with the field and DHMH laboratory data. The sediment program flags records that do not successfully merge with field and DHMH laboratory data and it is up to the data reviewer to resolve these discrepancies and ensure that sediment data are valid. Once all data are deemed valid, they are uploaded to DNR's Access water quality data base.

The final data set combining the field, lab, and sediment 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 data base directory for data user access. Data requests should be directed to Mark Trice, Program Chief of Water Quality Informantics (410-260-8630). A formatted submission data set and associated data documentation are 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 data base and prevents files that have fatal errors from being entered into CIMS. Non-fatal 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 data base, they are available to the public via the Bay Program data-hub (www.chesapeakebay.net). The data management process is diagramed in Figure 5.

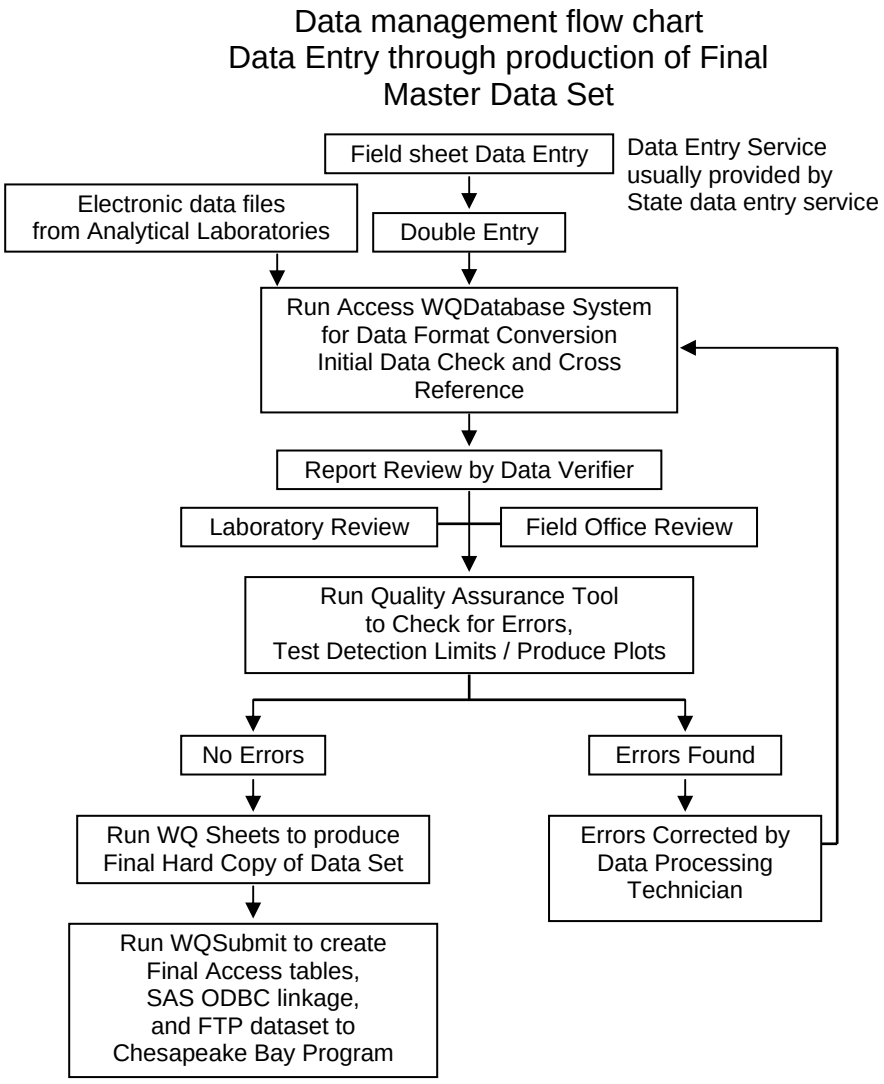


Figure 5. Data management flow chart

ASSESSMENT AND OVERSIGHT

Program and performance audits verify that procedures specified in the Project Plan are being followed. These audits ensure the integrity of the reported data.

C1 Assessments and Response Action

Field: 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 AMQAW and possible solutions are offered. The Field QA Officer regularly reviews Equipment Log Books to ensure that all staff are following QC procedures. Standard maintenance recommended by Hydrolab is performed at six-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.

Laboratory: Instrument preventive maintenance, repairs, and analytical corrective actions are documented on laboratory note books or the Division's Analytical Corrective Action forms. Corrective actions are initiated by the laboratory 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 (Appendix C) 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 3 or Chesapeake Bay Program Office staff.

Data Management: The Data Input Editor is the first line of defense for data correction. A DNR Administrative Aid reviews all incoming data and compares the data to the cross-reference file. The Administrative Aid verifies the submitted data and applies corrections to the physical data-sheet if errors are identified. During the data-import process, the Data Processing Programmer makes all corrections to the data and key fields as they are imported into the Water Quality Database System. The Data Processing Programmer 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 Water Quality 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

Reports to management are contained in the outline of deliverables in the project Scope of Work. Any changes to the QAPP or to the SOPs referenced herein will be documented and approval of the DNR Principal Investigator and Quality Assurance Officer will be obtained prior to implementation.

DATA REVIEW AND USABILITY

D1 Data Review, Verification, and Validation

These procedures are described in Section B10, Data Management and C1, Assessments and Response Action, above. The DHMH Environmental Chemistry Division uses data review checklists for data validation (Figure 6).

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 programmer who makes the changes (Figure 5).

D2 Verification Validation Methods

These procedures are described in Section B10, above.

D3 Reconciliation with User Requirements

Data collected under this program will initially be analyzed by staff of the USGS to calculate loads and estimate trends in nutrients and total suspended sediment using the minimum variance unbiased estimator developed by Bradu and Mundlak (1970) employing a seven parameter log-linear model (Cohn, et al., 1989). USGS ensures data quality by prescreening the input data through an examination of scatter plots. Preliminary output from the model is also screened for outliers by examining plots of residuals and box plots.

USGS developed the monitoring protocols and load and trend estimation techniques that will be employed to analyze data collected under this grant. These monitoring and data analysis techniques have been applied by USGS to data they collect under the River Input Monitoring Program, which started in the mid-1980s. As a result, the non-tidal network data should directly meet the objectives for which it is collected.

State of Maryland
DHMH - Laboratories Administration
DIVISION OF ENVIRONMENTAL CHEMISTRY
Nutrients Section

Figure 6. Data Review Checklist

LL Orthophosphate/EPA Method 365.1
LL Ammonia/ EPA Method 350.1

Lab Numbers[1]:_____

Date Collected: _____ Date Analyzed: _____ Analyst: _____

Procedure	Acceptance Criteria	Status *	Comments
Holding Time	48 hours @ 4°C; 28 days @ –20°C		
Samples Analyzed	Within 5 working days		
Calibration Curve	Corr. Coeff. $\geq$ 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 samples		
	Recovery = 90–110%		
External QC[2]	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 for OP; 0.008–0.500 ppm for NH_3)		
Diluted Samples	Correct final calculations		
Changes/Notes	Clearly stated		

REFERENCES

Bradu, D., and Mundlak, Y., 1970. **Estimation of log-normal linear models**. Journal of the American statistical association, 65(329), pp. 198-211.

Cohn, T.A., Delong, L.L., Gilroy, E.J., Hirsch, R.M., and Wells, D., 1989. **Estimating constituent loads**. Water resources research, 25(5), pp. 937-942.

Chesapeake Bay Program. 2008. **Nontidal Water Quality Monitoring. November 2008. Revised Chapter V of Recommended Guidelines for Sampling and Analysis in the Chesapeake Bay Monitoring Program, Revision 1-Draft**. Chesapeake Bay Program. Annapolis, Maryland. http://archive.chesapeakebay.net/pubs/subcommittee/msc/amqawg/Chapter%204%20Rev%201%20Mar%2026%2008.pdf

Quality Assurance Project Plan for the Maryland Department of Natural Resources Chesapeake Bay Water Quality Monitoring Program Chemical and Physical Properties Component for the period July 1, 2005 - June 30, 2006. Bruce Michael and Beth Ebersole

U.S. Environmental Protection Agency (EPA). 1993. **Methods for the Determination of Inorganic Substances in Environmental Samples**. (EPA/600/R-93/100).

U.S. Environmental Protection Agency (EPA). 1996. ***Recommended Guidelines for Sampling and Analysis in the Chesapeake Bay Monitoring Program***. Chesapeake Bay Program, August 1996. CBP/TRS 148/96; EPA 903-R-96-006.

U.S. Environmental Protection Agency (EPA). 1991. ***Chesapeake Bay Coordinated Split Sample Program Implementation Guidelines, May 1991***. Chesapeake Bay Program: Annapolis, MD. CBP/TRS 58/91.

Appendix A

Non-tidal Network Monitoring Program
Standard Operating Procedures

Maryland Department of Natural Resources

Non-tidal Network Program
Standard Operating Procedures

Prepared by: _____ Date: _____
Natural Resource Biologist

Reviewed by: _____ Date: _____
Natural Resource Biologist

Approved by: _____ Date: _____
Water Quality Monitoring Program Chief

July 2013

Table of Contents

MD-DNR
Non-tidal Network Program Protocols

Maryland's Non-tidal Water Quality Monitoring Network currently includes 21 load sites where nutrient and sediment concentrations are sampled monthly in addition to 8 times throughout the year during high flow events. A detailed description of sampling methods can be found in Chapter V (Non-tidal water quality monitoring) of the Analytical Methods and Quality Assurance Workgroup's *Draft Guidelines for Water Quality Sampling and Analysis*, which was approved by the Non-tidal Workgroup in November 2008 (http://www.chesapeakebay.net/committee_analyticalmethodsworkgroup_projects.aspx?menuitem=16701).

Load Sites: GEO0009, ANT0047, MON0546, BEL0053, TUK0181, WIL0013, NPA0165, GWN0115, DER0015, TF1.2, CAC0148, PXT0972, GUN0258, MKB0016, MGN0062, WCK0001, NWA0016, LXT0200, WIL0065, CVA0046, and 01636500. The sampling at gage number 0163500, Shenandoah at Millville, WV is performed by the U.S. Geological Survey (USGS).

Procedure: We will use a modified version of the USGS equal width interval assignment for our load sites. The PA USGS modified protocol reduces the number of verticals collected from 10 to a lower number based on the width of the stream being sampled.

Width of Waterway (ft.)	Minimum # of Verticals
0-25	1
25-100	3
100-250	5
250-500	7
>500	9

The number of verticals that will be used for MD-DNR sampling is dependent on the width of the stream at the time of sampling. The PA USGS modification assumes that the number of verticals required can be reduced because the stream is well mixed across the horizontal direction. To check this assumption each time a station is sampled four in-situ parameters (oxygen, pH, specific conductance and temperature) will be measured at each selected vertical sampling point. If the stream appears well mixed, the composite sample will be collected from vertical samples drawn at these selected sampling points. In addition, the variability across the stream will be checked every one to three years. This will be done by running the full nutrient suite on a sample generated at each vertical sampling point and on the normally generated vertical/horizontal composite. If variability is excessive, additional vertical sampling points (max 9) will be required for that station.

Once stream variability has been assessed, collection of a water sample that is both horizontally and vertically integrated will begin. If the maximum stream velocity observed is greater than or equal to 1.5 ft/sec and under 7.0 ft/sec an isokinetic equal width increment (EWI) composite sample will be generated using an approved USGS sampler (DH95 or DH81). If maximum stream velocity is under 1.5 ft/sec a non-isokinetic equal width increment composite sample will be generated using an approved USGS sampler (WBH-96 or modified DH81). At this time, MD

DNR does not have the capability to collect an isokinetic sample at sites with velocities over 7 ft/sec.

The EWI composite sample will be generated by collecting individual depth integrated samples at each specified vertical sampling point. These samples will be composited in either a 4L or 8L churn splitter. Two to three sample bottles will then be drawn from the churn splitter for field processing and /or delivery to the appropriate laboratory. One sample bottle will be processed in the field to generate both filter pads for the particulate nutrient parameters and a bottle of filtered sample water for the dissolved nutrient parameters. A second bottle will be filled for TSS analysis. All of the water in the TSS bottle will be used for the analysis. The TSS samples will be analyzed by the Maryland Department of Health and Mental Hygiene (DHMH). During storm events a third sample will be drawn and sent to the USGS Sediment Lab in Kentucky for suspended sediment analysis.

Please see the Hydrolab Sampling Procedures for a detailed explanation of how the in-situ parameters are sampled.

Please see Non-tidal Network Program Sampling Procedures for details on the collection of the EWI composite sample.

Please see the Non-tidal Network Program Sample Processing for a detailed explanation of how the field samples are processed.

Samples collected: Depth integrated samples collected at the vertical sampling points will be composited in the churn splitter. Sub-samples will then be dispensed from the churn splitter and used to generate:

One 500 ml HDPE bottle of whole water for Suspended Sediment Concentration analysis at the USGS Sediment Lab in Kentucky (during storm events and routine monthly samples that are impacted by high flow). Each quarter one sand/fine particle size analysis is done from the same bottle for each of the primary stations.

One TSS whole water bottle 20ml to 950 ml (volume dependent on turbidity)

One HDPE 2 quart nutrient sample bottle that will be field processed into:
- Two 25mm GF/F 0.7 micron Particulate Carbon/Particulate Nitrogen filters
- Two 47mm GF/F 0.7 micron Particulate Phosphorus filters
- Two 8 ounce HDPE bottles of filtrate for dissolved nutrient analysis

SAMPLE QA/QC

Duplicate stream samples are collected at a minimum of one (1) per water year per station. A total of twenty-four (24) duplicates are collected per water year. One (1) de-ionized water blank is processed per station per water year for a total of 20 de-ionized blanks.

Duplicate Sample

Twenty-four duplicate samples will be processed per water year. Each sample site has a minimum of one duplicate sample per water year. Sixty percent (60%) of the duplicates will be collected as routine samples. The remaining forty percent (40%) will be collected during storm sampling events. This breakdown of duplicate sampling mimics the percentages that each site

is sampled per water year. The duplicate will be drawn from the same churn splitter as the original sample and processed identically. See Churn Splitter Sub-Sampling Procedure and Non-tidal Network Program Sample Processing for a detailed explanation of these processes. Results will be reviewed as part of MD DNR duplicate data set.

DI Blanks: Processed and Unprocessed

One DI water equipment blank is processed and submitted for each sample site, once per water year. Sixty percent (60%) of the DI blanks will be collected as routine samples. The remaining forty percent (40%) will be collected during storm sampling events. This breakdown of DI Blank sampling mimics the percentages that each site is sampled per water year. **Procedure:** Rinse the appropriate sample bottles with DI water; use the type of sample bottle that will be used at the station you are currently visiting, under the current conditions. Rinse the appropriately-sized churn splitter (for the current station & conditions) thoroughly with DI water. Fill the sampling bottle with DI water and pour into the rinsed churn splitter. Actively churn the DI water sample according to the Churn Splitter Sub-Sampling Procedure. Collect one bottle for SSC (only if doing a DI Blank during Storm Event Sampling), one bottle for TSS and one 2 quart bottle to be processed for particulate and dissolved nutrients. Process this bottle in the same way that stream samples are processed. Generate filtrate, PC/PN filters and PP filters. Also submit an unprocessed blank to be analyzed for dissolved nutrients. This whole water sample is taken from the supply of DI water that is used for the processed DI water blank. The unprocessed sample should be submitted in two 8 ounce bottles, labeled “whole water, 1 of 2 and 2 of 2”. Ice samples and ship with regular stream samples. Results will be reviewed by the Field Office Chief. Problems will be discussed with the Project Officer and potential solutions explored.

Routine Sampling Frequency

Both primary and supplemental stations are sampled once per month on a predetermined schedule, these are our Routine Samples (RS).

If high discharge occurs during routine monthly sampling, samples are collected on the scheduled date using procedures for storm event sampling, including a SSC sample (primary stations only). These samples are to be counted as routine, monthly samples and designated as sample type “Routine, Storm-impacted” (RI).

Storm Sampling Frequency

Eight storm event samples per station are required each year, with an attempt to collect 2 storm events per quarter.

A storm event has a rising discharge (cfs) that is a significant change above the average daily discharge with a minimum of two times the pre-storm discharge value. The storm event triggers are site specific and are usually a five- to ten-fold increase over the average daily discharge. Samples are collected at any point in the hydrograph, i.e., rising or falling limb, or at peak discharge. Two samples may be collected during a single storm event, however these samples must be collected on different days. This also applies to storm sampling following the collection of a routine, storm-impacted sample.

Non-tidal Network Program Sampling Procedure

1. You will need to record the time and gage height at the beginning and end of the sample collection period on the field sheet. If you are sampling at the exact location of the gage, open the gage house and record the gage height and time before sampling and after you finish collecting samples. If you will not be sampling at the exact gage location stop at the gage and record a beginning gage height and time. If the gage is on-line you can get the end reading from the USGS web page. Check the recorded gage height against the cross reference list in the field pack to determine if maximum velocity expected is greater than or equal to 1.5 ft/sec. **If YES,** an isokinetic composite sample **MUST** be collected. If the maximum velocity is under 1.5 ft/sec or over 7.0 ft/sec a non-isokinetic composite sample is collected.

2. Set up cones on the road to block off enough room so that you feel safe on the downstream side of the bridge. Wear your orange safety vest.

3. Measure stream width by placing the measuring tape along the bridge from stream bank to stream bank. Measure from left to right looking downstream. Establish the number of increments (transects) you will sample by using the table on page 3. After you determine the number of increments (transects) that will be sampled, use the formula below to determine the location of each vertical sample. A more detailed explanation of the new isokinetic sampling protocols can be found in the Non–tidal Field Pack. Check "Sampling Procedures and Protocols for the Chesapeake Bay Non-tidal Water Quality Network" report, page 6 or the USGS procedures.

   Here is a formula to determine the number of transects and the location of verticals:

   - Stream Width / number of transects = transect length
   - 1^{st} vertical = Transect length / 2
   - 2^{nd} vertical = Transect length + 1^{st} vertical
   - 3^{rd} vertical = Transect length + 2^{nd} vertical
   - 4^{th} vertical = Transect length + 3^{rd} vertical
   - 5^{th} vertical = Transect length + 4^{th} vertical

   For example, if the stream is 60 feet wide and it is divided into 3 transects, the 1^{st} sample should be taken at 10 feet from the left bank (while facing downstream), the 2^{nd} at 30 feet, the 3^{rd} at 50 feet. Record vertical locations in the comment section of the field sheet.

4. Once you have established a location for each vertical, record Hydrolab readings for each vertical by immersing the Hydrolab in the stream directly (if equipment and stream velocity are suitable) or by collecting a rinsed bucket at each vertical. You must note on the field sheet if the readings were taken from bucket samples. Refer to Hydrolab Sampling Procedures for a detailed explanation of this process. Review these readings. The stream is well mixed if no set of readings for any one parameter differs by 20 %. If the stream is well mixed, record the median value for each parameter on the field sheet and begin collecting samples. If stream is not well mixed, increase the numbers of verticals by at least two and repeat steps 3 & 4.

5. Based on the stream velocity and sampler choices available, choose a sampler to use. Rinse the sampler collection bottle and all whole water sample bottles three times with stream water (directly in stream or from a freshly collected bucket of stream water). Rinse the churn splitter with 2 to 4 L of stream water. Run a liter of water through the spigot.

1) **DH-95** (bronze, plastic coated isokinetic sampler)

A. Follow instructions 1 – 5 under Non-tidal Network Program Sampling Procedure.

B. Connect the sampler to a hanger bar and the hanger bar to the suspension cable on the crane or the bridge board.

C. Decide who will be the clean hands person (who handles the sample) and who will be the dirty hands person (who handles the equipment). The clean hands person must wear rubber gloves during the sampling. They will only touch the sample bottle and churn splitter.

D. Select the largest diameter nozzle size that the transit rate and depth will allow. The largest size (5/16" diameter) nozzle is the most commonly used, and seems to be appropriate for most situations. Screw the selected nozzle into a clean cap and bottle configuration.

E. Lift the o-ring and place the bottle configuration into the sampler cavity. The o-ring should fit over the neck of the bottle and hold it in place. Rotate the bottle until the air vent hole is vertical. Visually check the nozzle and air vent hole for obstructions.

F. If the stream looks deeper and faster in one area, establish your transit rate at that spot before starting to sample. Remember, if the sampler collection bottle is over-filled (more than ¾ full) at any of the verticals using the transit rate you establish initially, you must discard all sample water in the churn splitter and begin sampling over again. The sampler collection bottle must still be bubbling when it reaches the surface. If the collection bottle is less than 40% filled after the first up/down pass, you can lower and raise it again before you empty it into the churn splitter. Remember total amount collected must be ¾ or less of the liter bottle. **If you decide to do multiple dips, you must do the same number of dips at each vertical sampled.**

G. Once you establish your transit rate and number of dips, you are ready to collect the first vertical. Lower the DH-95 sampler until the tail makes contact with the water surface. Record the number from the reel in the "Cable reading - start" line on the field sheet.

H. Wait until the sampler orients towards the flow before lowering the sampler into the water column. Once it has aligned, begin to lower the sampler at a fixed rate, when you feel it touch the bottom, quickly note the number on the reel and reverse directions to raise the sampler to the surface. Record the number from the reel in the "Cable reading - end" line on the field sheet. Repeat, if doing multiple dips.

I. Raise the collected sample. The clean hands person should then retrieve the bottle from the sampler and empty it into the churn splitter. Gently swirl the bottle to ensure that all sediment has been transferred to the churn splitter.

J. Move the sampler to the next vertical location. Repeat steps G through I for each vertical.

K. Continue this process until the verticals for all transects have been completed. The churn splitter must contain enough water to process all the samples. If necessary you can return to each vertical location and collect an additional sample if more water is needed. Remember, the churn splitter cannot be overfilled!!! If it does overfill, you must empty the churn splitter and begin the sampling over again.

L. Follow the instructions under Churn Splitter Sub-Sampling Procedure at the end of this section.

2) WBH-96 (Weighted bottle, hand held, non-isokinetic sampler)

A. Follow instructions 1 – 5 under Non-tidal Network Program Sampling Procedure.

B. Put the plastic liter size collection bottle in the sampler and place the elastic around the neck of the bottle making sure it is secure.

C. Decide who will be the clean hands person (who handles the sample) and who is the dirty hands person (who handles the equipment). The clean hands person must wear rubber gloves during the sampling. They will only touch the sample bottle and churn splitter.

D. If the stream looks deeper and faster in one area, begin there. Remember, if the sample collection bottle is over-filled (past the neck of the bottle) you must discard the sample water and begin again. The sample collection bottle must still be bubbling when it reaches the surface.

E. Lower the sampler until the bottom of the sampler is touching the water surface. Begin lowering the sampler and when you feel it touch the bottom, automatically begin to raise the sampler to the surface.

F. Raise the collected sample. The dirty hands person should set the sampler on the ground and the clean hands person should then retrieve the bottle from the sampler and empty it into the churn splitter.

G. Continue this process until the verticals for all transects have been completed. The churn splitter must contain enough water to process all the samples. If necessary you can return to each vertical location and collect an additional sample if more water is needed. Remember, the churn splitter cannot be overfilled!!! If it does overfill, you must empty the churn splitter and begin the sampling over again.

H. Follow the instructions under Churn Splitter Sub-Sampling Procedure at the end of this section.

3) DH-81 (Hand held wading sampler, optional isokinetic/ non-isokinetic)

If stream velocity is ≥1.5 ft/s and considered safely wadable, you may use the DH-81 as an isokinetic sampler by using the appropriate nozzle (usually 5/16”). If flows are less than 1.5 ft/s the DH-81 may be used without the nozzle to obtain a grab sample.

A. Select the area of stream that you will be sampling and secure the tape measure across the stream. Measure from left to right looking downstream. Establish the number of increments (transects) you will be sampling and location across the transect where you will be collecting your vertical sample based on the width of the stream.

B. Once you have established a location for each vertical, record Hydrolab readings for each vertical. Have one person on the stream bank recording the numbers while the other person handles the Hydrolab. See Hydrolab Sampling Procedures for a detailed explanation of the process.

C. Rinse the DH-81, including bottle, nozzle (if needed), cap and the churn splitter in the stream. Make sure you are downstream of the sample area to ensure that you do not stir up the streambed prior to sampling.

D. Assemble the DH-81 by screwing the cap onto the liter sample bottle and attach the nozzle to the cap (if the steam velocity is under 1.5 ft/sec you can sample without the nozzle). Secure the DH-81 to the wading rod by snapping it into place over the cap.

E. Decide who will be the clean hands person (who handles the sample) and who is the dirty hands person (who handles the equipment). The clean hands person must wear rubber gloves during the sampling. They will only touch the sample bottle and churn splitter.

F. Enter the stream down river of the sampling location and walk up to the sampling location in the centroid (maximum) of the steam flow. Raise and lower the sampler at a constant rate such that the sample bottle is ½ - ¾ full when breaking the surface.

G. **If the sample bottle is too full pour out the sample and speed up your transit rate or use a smaller nozzle or a combination of both until the sample bottle fills ½ - ¾ when the sampler is raised out of the water column. Likewise, if the sample is not full enough, pour out the sample and use a larger nozzle or slow your transit rate to increase sample volume.**

H. Empty the collected sample into the churn splitter. Move to the next vertical and repeat the collection process.

I. Repeat the sample collection process until there is sufficient volume to fill the 4 or 8 Liter churn splitter.

J. Follow the instructions under Churn Splitter Sub-Sampling Procedure at the end of this section.

4) DH-59 (Brass, hand-held isokinetic sampler with a fixed nozzle.)
As of December 2005, the DH-59 has not been used. Refer to a previous revision for instructions regarding the use of the DH-59.

5) Bucket Sampling Note: If D.O. and Temp are read from a bucket sample YOU MUST Enter a B in the G/L box associated with D.O. so that these values are deleted from data sent to CBP.

Bucket samples are taken from bridges. A sample may be collected to provide stream rinse

water for sampling equipment and whole water bottles. If in-situ readings cannot be made by immersing Hydrolab directly in the stream you can collect a bucket from each vertical point for readings. See note above.

A. Select the appropriate length of rope for the bridge from which you will be sampling and secure tightly to bucket.

B. Chose a vertical sampling location to sample.

C. Lower the bucket to the water.

E. Tip the bucket and fill with enough water to rinse the bucket (at least a few inches).

F. Depending if it is a high or low 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. Rinse three times.

G. Fill the bucket.

H. 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.

I. Immediately carry the bucket back to the van. Rinse equipment or go to J.

J. If using for in-situ readings immerse the Hydrolab sonde in the bucket, swirl at 1 ft/sec. Record readings. Repeat for all verticals. Remember to record a “B“ in G/L box for D.O.

Churn Splitter Sub-Sampling Procedure

The following steps are to be completed for filling of all sample bottles from the churn splitter:

A. Set the churn splitter in an area where the spigot is easily accessed to dispense water.

B. One person should churn the sample, while the other person fills the sample bottles from the churn splitter.

C. Churn the sample at 9 inches per second.

D. Do not break the water surface with the wand while churning.

E. Churn the sample a minimum of 10 times before dispensing water.

F. Continue to churn the sample until all the sample bottles are filled. Samples cannot be dispensed if the water level is at or below the spigot.

G. Dispense whole water sediment related samples first. Dispense:
   a. SSC (storms only)
   b. TSS
   c. Nutrient Filtration Bottle

   Note: After filtering for PC/PN if the sample volume originally chosen for the TSS sample is too small or too large, discard and dispense a TSS bottle with a better volume.

H. Process sample bottles filled as per the instructions under Non-tidal Network Program Sample Processing.

Churn Splitter Cleaning Procedure

After all samples are collected and processed empty churn splitter and rinse well with DI water. Rinse sampler collection bottle with DI. If any of the sample collection equipment needs to be reused before it can be cleaned at the office, follow procedure below.

A. Soak equipment in 10% Liquinox Solution for 20 – 30 mins. If churn splitter is being cleaned fill it with Liquinox and add collector bottle and nozzles. Let sit while completing station or while driving to next station. There is a small cup for soaking just nozzles in the field tub.

B. After soaking, scrub with the brush provided and rinse completely with tap water. Rinse three times with DI. Air dry and store in clean baggies or use again.

Non-tidal Network Program Sample Processing

A. Laboratory Supplies

Pads

a) PC/PN
The pads used for PC/PN samples come directly from DHMH. The PC/PN pads are pre-combusted (490 ^{o}C), 25mm Whatman GF/F glass fiber filters – pore size 0.7 µm. Two PC/PN pads are used per sample.

b) PP
The pads used for PP samples are 47mm Whatman GF/F glass fiber filters - pore size 0.7 µm. Two PP pads are used per sample.

B. Particulate sample filtration, processing and storage

1. **Particulate Carbon/ Particulate Nitrogen (PC/PN)**
   a) To generate PC/PN filters first clean two 25mm bells 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.
   b) Place a pre-combusted 25 mm GF/F filter (pore size = 0.7 µm) on each filter frit. Always use clean forceps when handling the filter pads.
   c) Using the two quart whole water sample drawn from the churn splitter **(please see Section D- Churn Splitter Sub-Sampling Procedure)** mix sample thoroughly by agitating and shaking the sample bottle vigorously, then rinse graduated cylinder three times with sample.
   d) 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 inches of Hg is good).
   e) Filter 10-200 ml through each filter. Filter enough sample to leave noticeable color on the filter pad.
   f) Make sure filter is sucked dry and the **same volume is filtered for both pads**.
   g) 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.

   h) Using forceps, fold each filter in half.
   i) Place both filters in a foil square labeled with date, station, sample layer, PC/PN, and volume filtered. Be sure that the pads are not overlapping in the foil square to keep them from freezing together.
   j) 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. Place the folded foil in a zip-lock bag or pad container, and put it in a cooler on ice.

k) Upon return to the Field Office, place the foils in their appropriate zip-lock bag in the sample freezer and place the bag in the DHMH bin. Put the completed volume sheet in the bag with the foils.

2. **Particulate Phosphorus/ Particulate Inorganic Phosphorus (PP/PIP)**

a) To generate PP filters, clean two 47mm bells 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. The filters used are two Whatman 47 mm GF/F filters.
b) Using the two quart whole water drawn from the churn splitter filter 50 ml of sample through each filter pad.
c) Use the filtrate as an equipment rinse and discard.
d) Then filter enough additional (another 50 - 750 ml) to leave a noticeable color on the filter pad.
e) 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.
f) Use this filtrate to fill up the container for the dissolved parameter analysis. See section C (Filtered dissolved nutrient sample collection) below.
g) After collecting filtrate, make sure filter is sucked dry.
h) Rinse the filter pad using at least three - 10 ml rinses of DI water sucking the pad dry after each rinse.
i) Using forceps, fold each filter in half.
j) Place both filters in a foil square labeled with date, PP, station, sample layer, 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.
k) Fold the foil square as described in step B.1.i., above. Place foil square in zip-lock bag or pad container, and put in the cooler on ice until you return to the field office.
l) Upon return to the Field Office, place the foils in their appropriate zip-lock bag in the sample freezer and place bag in the DHMH bin. Put the completed DHMH volume sheet in bag along with the foil squares. Frozen samples are delivered Friday of sampling week so the Lab can analyze them within 28 days of collection.

C. Dissolved nutrient sample filtration, collection and storage

NOTE: The filtrate collected for this sample must come from the PP filtration set-up. If you cannot get enough water through these pads to fill the filtrate sample bottle, then use more GF/F filters to get enough filtrate. The filtrate may not come from pads that are pre-combusted (PC/PN).

The following steps are to be completed for collection of all filtrate:
a) Run 50 ml of sample water through the filter.
b) Use this 50 ml of filtrate to rinse the flask and then discard.
c) Run more sample water through the filter and collect in the flask.
d) Label two 8 ounce HDPE bottles with “NTN”, station, date, “Filtrate” and “1 of 2” on the first bottle and “2 of 2” on the second bottle.
e) Rinse the two labeled 8 ounce HDPE bottles and caps three times with filtrate.
f) Fill the bottles with filtrate and replace cap. If sample will be frozen before delivery to the lab do not fill more than ¾ full.

g) Store the bottle on ice in a cooler. Deliver to the courier or directly to DHMH at the end of the field day. If you miss the courier filtrate sample bottles may be frozen and delivered on Friday directly to DHMH. If freezing the filtrate sample, copy the regular DHMH Lab Sheet and place the copy in a zip-lock bag with the filtrate bottle. Lab must analyze unfrozen sample within 24 hrs of collection; frozen sample within 28 days.

D. Total Suspended Solids (TSS) collection and storage

a) Choose the appropriate size sample bottle.** Label with "NTN", Station Id, date and "TSS ONLY". Rinse cap and bottle three times with sample water. Bottle may be rinsed from a Rinse Only bucket of water.

b) Fill bottle with sample from the churn splitter. Theoretically the TSS bottle should be filled before the nutrient bottle. Follow the Churn Splitter Sub-Sample Procedures. Because DHMH will be sampling the entire amount we send them, you cannot dump any water out once you fill the container.

c) Ice sample. Deliver directly or send by courier to the DHMH within 72 hrs. An original completed DHMH Lab Sheet must accompany the sample.

Note: If you are sampling on a weekend, or miss the courier, the holding time for TSS samples is one week. The sample can be sent by courier the day after you collected it OR delivered directly to the Lab within 72 hours of collection.

** Note: DHMH will be using all the water in the TSS sample bottle to generate the TSS filter. DO NOT SEND THEM TOO MUCH WATER. The DHMH uses 47 mm filters for TSS. Fill the TSS Sample Bottle with the same amount of water that you expect to use for the PC/PN filters.

REMEMBER the Lab will need to rinse their TSS filters THREE TIMES. There are 60 ml, 8 oz. (237 ml), 16 oz. (473 ml) and 1 qt. (947 ml) bottles in the field tub to use for the TSS sample. The 60 bottles are graduated. The 8 oz,16 oz and 1 qt bottles can be filled to any estimated volume.

E. Suspended Sediment Concentration (SSC) collection and storage

a) Label 500 ml plastic Nalgene bottle with Station ID, date and time.

b) Fill bottle to shoulder from churn splitter. Follow the Churn Splitter Sub-Sample Procedure instructions. No water can be dumped from the filled Nalgene bottle. **This sample must be the first sample taken from the churn splitter.**

c) Mark water level line on the Nalgene bottle with a Sharpie.

d) Sample does not need to be iced but should be kept in the dark. Place in the box labeled "Sediment Samples" when you return to the office. Samples will be shipped quarterly to the USGS Sediment Lab in Kentucky. Note: Each year (Oct-Sept.) all samples must be shipped by September 30th.

Hydrolab Calibration Procedures

A. All instruments are calibrated prior to field use. Calibration is typically completed the Friday prior to the sampling week. If office time is available on a closer day preceding the run, then the calibration will be completed at that time.

B. When large adjustments to the dissolved oxygen (DO) value were necessary during calibration, the DO should be checked prior to taking the instrument in the field. Instrument calibrations may be also be rechecked in the calibration lab any time the field readings seem suspect.

C. The DO is checked in the morning prior to sampling when significant changes in barometric pressure have occurred. This is typical of the Western Maryland NTN run.

D. Instruments are post-calibrated (checked for their accuracy) 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.

I. Series 3 Instrument

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 an array of 6 nickel electrodes oriented in two vertical rows of three with each row inside adjacent parallel channels of a standard plastic block. Individual electrodes are oriented horizontally. The conductivity reading is corrected to standard temperature. 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 set 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 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.

II. Series 4a Instrument

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. 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.

III. Series 5 Instrument

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. 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.

Hydrolab Sampling Procedures

Make sure the Hydrolab has been turned on for at least 15 minutes prior to sampling. Submerge the Hydrolab sonde directly in the stream to obtain the required readings. If flow is too swift or your meter is not practical for in-situ readings obtain readings from a bucket grab. Add B in G/L box for DO and note “Bucket Readings” in “Comments” on Field Sheet.

1. Remove plastic storage cup from the Hydrolab, check DO membrane to make sure it is clear and there are no bubbles.

2. Protect probes by installing probe guard or stirrer if flow will be under 1 foot per second for any in stream measurement point .

A. Sampling in the bucket: (Remember “B” in DO G/L box

a) Follow steps 1 and 2 above.
b) Swirl the Hydrolab in the bucket at 1 foot per second until the readings stabilize.
c) Record readings on the field sheet.
d) Remove the Hydrolab from the bucket, and rinse probes with de-ionized water before replacing plastic storage cup.

B. Sampling from the bridge:

a. Follow steps 1 and 2 above.
b. Turn on the circulator from the Hydrolab display menu.
c. Lower the Hydrolab over the bridge at the first vertical. Position the probes at mid-depth for the vertical.
d. Wait for the readings to stabilize and record them on the field sheet.
e. Carefully raise the Hydrolab back up and move to the next vertical.
f. Repeat steps c through e until all the verticals have been sampled.
g. Remove guard. Rinse probes with DI. Replace storage cup.

C. Sampling while wading:

a) Follow steps 1 and 2 above.
b) Turn on the circulator from the Hydrolab display menu.
c) Place the Hydrolab directly in the water at the first vertical. Position the probes at mid-depth for the vertical.
d) Wait for the readings to stabilize and record them on the field sheet.
e) Repeat steps c and d until all the verticals have been sampled.
f) Remove guard. Rinse probes with DI. Replace storage cup.

YSI ROX Calibration Procedures

All instruments are calibrated prior to field use. Calibration is typically completed the Friday prior to the sampling week. If office time is available on a closer day preceding the run, then the calibration will be completed at that time.

When large adjustments to the dissolved oxygen (DO) value were necessary during calibration, the DO should be checked prior to taking the instrument in the field. Instrument calibrations may be also be rechecked in the calibration lab any time the field readings seem suspect.

The DO is checked in the morning prior to sampling when significant changes in barometric pressure have occurred. This is typical of the Western Maryland non-tidal network run.

Instruments are post-calibrated (checked for their accuracy) 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.

ASTM D888 – 09 (C) Calibration

Add a sufficient amount of reagent grade water to a bucket to adequately cover the ROX probe. Allow the water to equilibrate to room temperature ($\pm$ 2^{o}C). Using a steady stream of clean compressed air (approximately 10 to 40 mL per minute flow rate) aerate the water for a minimum of 30 minutes. Allow the water to re-equilibrate to room temperature ($\pm$ 2^{o}C) for 45 to 60 minutes. Note the barometric pressure and sample temperature and use the values to calculate the theoretical dissolved oxygen concentration from a dissolved oxygen table. Analyze the dissolved oxygen concentration immediately if unable to seal the top of the bucket. Provide for suitable turbulent flow past the sensor cap. Verify the calibration by taking a second reading. The calibration verification should be within 97 to 104% of the theoretical dissolved oxygen concentration. If the calibration verification is outside of the theoretical recovery range, re-calibrate the sensor. For a two point calibration, these procedures should be followed for the saturation point and the zero calibration point should be verified.

Log of Significant Changes for the Non-Tidal Network Program

Date Initiated	Procedural Changes
January 1, 2005	The non-tidal network sampling program for base flow and storm events was initiated. Samples were analyzed by the Maryland Department of Health and Mental Hygiene using whole water analyses and concurrently by the Chesapeake Biological Laboratory using filtered water analyses. Both laboratories analyzed samples through June 2005.
July 1, 2005	The Maryland Department of Health and Mental Hygiene laboratory initiated filtered water analyses for all laboratory parameters except total suspended solids. Due to concerns about potential methods and laboratory change effects, data for January through June 2005 were not delivered to the Chesapeake Bay Program. Data collected from July 1, 2005 onward have been submitted to the Bay Program data base. As of July 2005 the program consisted of the following ten stations: ANT0047, BEL0053, DER0015, GEO0009, GWN0115, MON0546, NPA0165, TF1.2, TUK0181, and WIL0013. The stations were sampled during base flow and storm events and were classified as primary stations in the non-tidal network.
October 1, 2005	Three stations (GUN0258, PXT0972, and CAC0148) were added to the network, but only sampled during base flow conditions. These stations were classified as secondary stations in the non-tidal network.
November 1, 2005	Discontinued use of the DH-59. After this point, all samples were collected with one of the following: DH-95, DH-81 or WBH-96.
January 1, 2007	GUN0258, PXT0972, and CAC0148 were upgraded to primary sites and sampled during base flow and storm event conditions.
July 1, 2008	Began collecting Routine Impacted samples. Until July 2008 all sites that were influenced by storms were collected as storm event samples. All routine stations were sampled under “baseflow”. If a storm occurred during a scheduled baseflow run, a storm sample was collected and the baseflow (routine) was rescheduled for collection another day.
July 1, 2009	The Department of Health and Mental Hygiene Laboratory switched from using a 24 millimeter diameter Whatman Grade 934-AH: 1.5 µm filter pad to an 47 mm diameter 1.5 µm Environmental Express filter pad for the analysis of total suspended solids.
March 25, 2010	The Department of Health and Mental Hygiene Laboratory changed analysis methods for ammonia from the phenate method to the salicylate method. Differences in the analytical results for the two methods were assessed in the Data Analysis Issues Tracking System report #049 which showed that the salicylate method yielded slightly higher results than the phenate method ($p<0.0001$). DHMH switched back to the phenate method on November 5, 2010.
October 17, 2011	Sampling was initiated at MKB0016 on Manokin Branch.
October 24, 2011	Sampling was initiated at MGN0062 on Morgan Creek.

Log of Significant Changes for the Non-Tidal Network Program

(continued)

Date Initiated	Procedural Changes
October 25, 2011	Sampling was initiated at NWA0016 on Northwest Branch Anacostia River and LXT0200 on Little Patuxent River.
October 27, 2011	Sampling was initiated at WCK0001 on Wheel Creek.
	Three stations were added in the spring of 2012 using funding from the 2010 Chesapeake and Atlantic Coastal Bays Trust Fund. These stations include CVA0046, WIL0065, and at USGS gage number 01636500 near Millville, WV.

Appendix B

Cross Reference Sheet
Documentation and Procedures

Cross Reference Sheet Procedures

The following documentation outlines the conventions for filling out the cross reference sheet. (Note: Although this sheet is labeled "Progress Report," it is actually the Cross Reference Sheet) The cross reference sheet is sent along with field sheets from the field office to the DNR Tawes Building, so that the DNR data management staff knows what data to expect in the form of field sheets and laboratory data.

The cross reference sheet includes the name of the program, the sampling month and year, the name of the field office representative who originates the sheet, the station name, the sampling day, the sampling depth, the sequence number, layer code, and columns for tracking laboratory sheets and if a sediment sample was collected for analysis at the USGS sediment laboratory. There is also a comment line to explain missing samples, stations, field abnormalities, sampling intervals, or potential data problems.

1. The name of the program
2. The sampling month and year
3. The name of the Field Office representative who originates the sheet
4. The station name
5. The sampling day
6. The sequence number
7. The sampling depth
8. The layer code
9. Column for tracking receipt of laboratory data
10. Column for USGS sediment sample taken
11. Column for comments

An example Cross Reference Sheet (labeled "Progress Report/Cross Reference Sheet") follows.

Maryland Department of Natural Resources MANTA
Chesapeake Bay Water Quality Monitoring Progress
Report - Non-tidal Network Baseflow Sampling

Station	Day	Sequence Number	Sample Depth	Layer code	DHMH	USGS sediment	Comments
GEOOO09	26	0707801	Start 0.0 End 0.2	VH	0185	N/S	26 feet total. 3 verticals @4.4, 13 and 21.7 feet. Waded at gage. Water very dark. Scattered thunderstorms last evening. Water depths (M): I (0.4),2 (0.3), 3 (0.2)
ANT0047	25	0707802	Start 0.0 End 0.4	VH	0179	N/S	92 feet total. 7 verticals @ 8 (eddy - 6.6), 19.6,32.8,45.9,57 (eddy - 59), 72.2 and 82 (shallow and rocks at 85.2ft) feet from the left. Sampled from the bridge. Meter K's DO membrane had a hole in it after this station, and was not able to be post- calibrated. Water depths (M): I (0.4),2 (0.7), 3 (1.0),4 (1.1), 5 (0.9),6 (0.4), 7 (0.3)
MON0546	25	0707803	Start 0.0 End 0.2	VH	0181	N/S	79 feet total. 5 verticals@ 7.9, 23.7, 39.5, 55.3 and 71.1feet from the left. Sampled from the bridge. Non-isokinetic sample. Extremely low water level. Lots of algae on rocks. Meter K's DO membrane had a hole in it after doing the Antietam station, and was not able to be post- calibrated. Water depths (M): 1 (0.2),2 (0.4), 3 (0.4),4 (0.5), 5 (0.2)
TUKOl81	2	0707804	Start 0.0 End 0.9	VH	0005	N/S	96 feet total width. 5 verticals @ 9.6, 28.8, 48, 67.2 and 86.4ft from the left. Sampled from the bridge. Non-isokinetic sample. Rinsed PC/PN pads with 200m I of D I before filtering sample. Water depths (M): I (0.7),2 (1.6), 3 (2.7),4 (1.8), 5 (2.0).

Appendix C

Corrective Action Form for the Department of Health and Mental Hygiene

State of Maryland
DHMH - Laboratories Administration
Division of Environmental Chemistry

DIVISIONAL ANALYTICAL CORRECTIVE ACTION FORM

Quality Assurance Program

■ NONCONFORMANCE

Customer:		Samples(s):	
Test:	Method:	Instrument:	Date of Occurrence:

☐ Failed Tuning ☐ Failed Calculation ☐ Instrument Instability ☐ Instrument Malfunction ☐ Other	☐ Power Failure ☐ Broken or Lost Aliquot ☐ Insufficient Volume ☐ Poor Aliquot Preservation	☐ Exceeded Holding Time ☐ Matrix Interference ☐ Out-of-Control QC

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:

■ ACKNOWLEDGEMENT

Signature of Division QA Officer:	Date:

Copies: Division QA Officer
Laboratory File

DEC/QA9
8/2007

Appendix D

Standard Operating Procedures for the Department of Health and Mental Hygiene

State of Maryland
DHMH - Laboratories Administration
DIVISION OF ENVIRONMENTAL CHEMISTRY

STANDARD OPERATING PROCEDURES

Title:	Determination of Total Dissolved Nitrogen Flow Injection Colorimetric Analysis EPA Method 353.2
SOP No.:	IAL-SOP-EPA 353.2/R2.0-11
Revision:	2.0 Replaces: 1.0 Effective: 8/18/2011
Laboratory:	Inorganics Analytical Laboratory
POC:	Shahla Ameli amelis@dhmh.state.md.us

Laboratory Supervisor: _____ _____
Signature Date

Division's QA Officer: _____ _____
Signature Date

Division Chief: _____ _____
Signature Date

State of Maryland
DHMH - Laboratories Administration
DIVISION OF ENVIRONMENTAL CHEMISTRY
201 W. Preston Street
Baltimore, MD 21201

REVISION RECORD

Revision	Date	Changes	Made By	Effective Date
0.0	6/3/08	N/A	Shahla Ameli	6/2/08
1.0	12/09	Screen snapshots added to procedure, new Sample Run Log, tracking IDs for standards and reagents	Shahla Ameli	1/10
2.0	8/11	New SOP tracking number, technical and editorial changes	Shahla Ameli	8/18/11

TABLE OF CONTENTS

STANDARD OPERATING PROCEDURES

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 then 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.

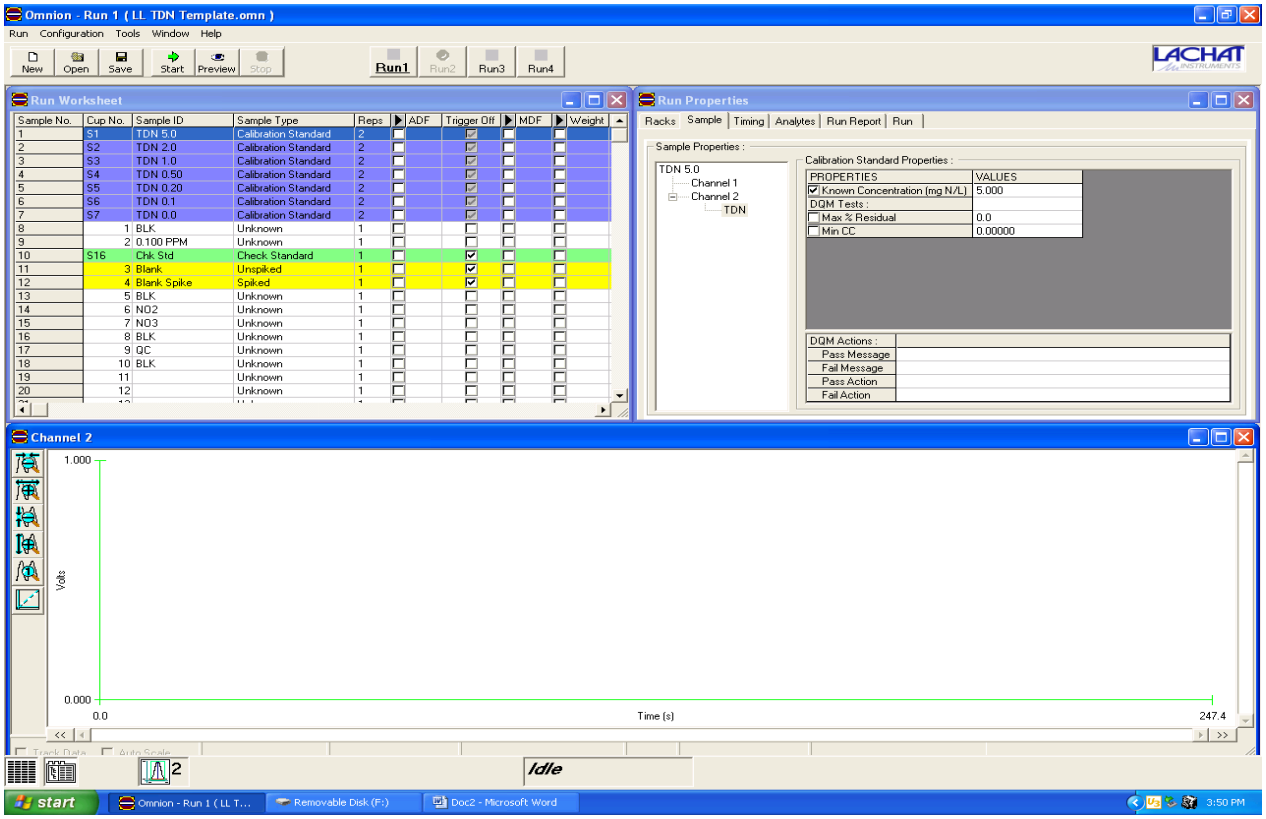
9.2.7 Get the data reviewed by a designated scientist, and then, report the results on the Analysis Request Forms.

9.3 Instrument set-up and sample analysis

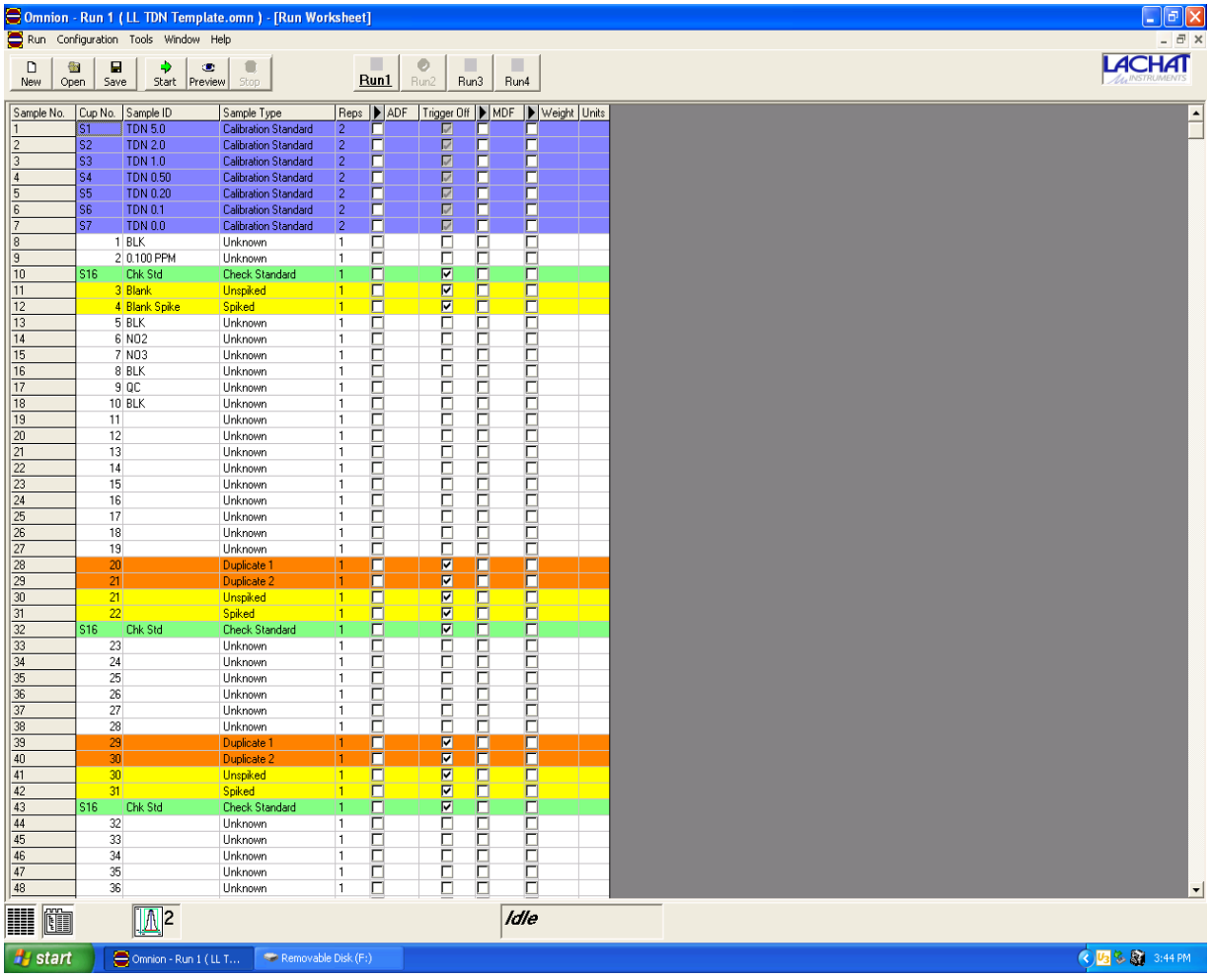
9.3.1 Set up manifold as in the diagram.

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.



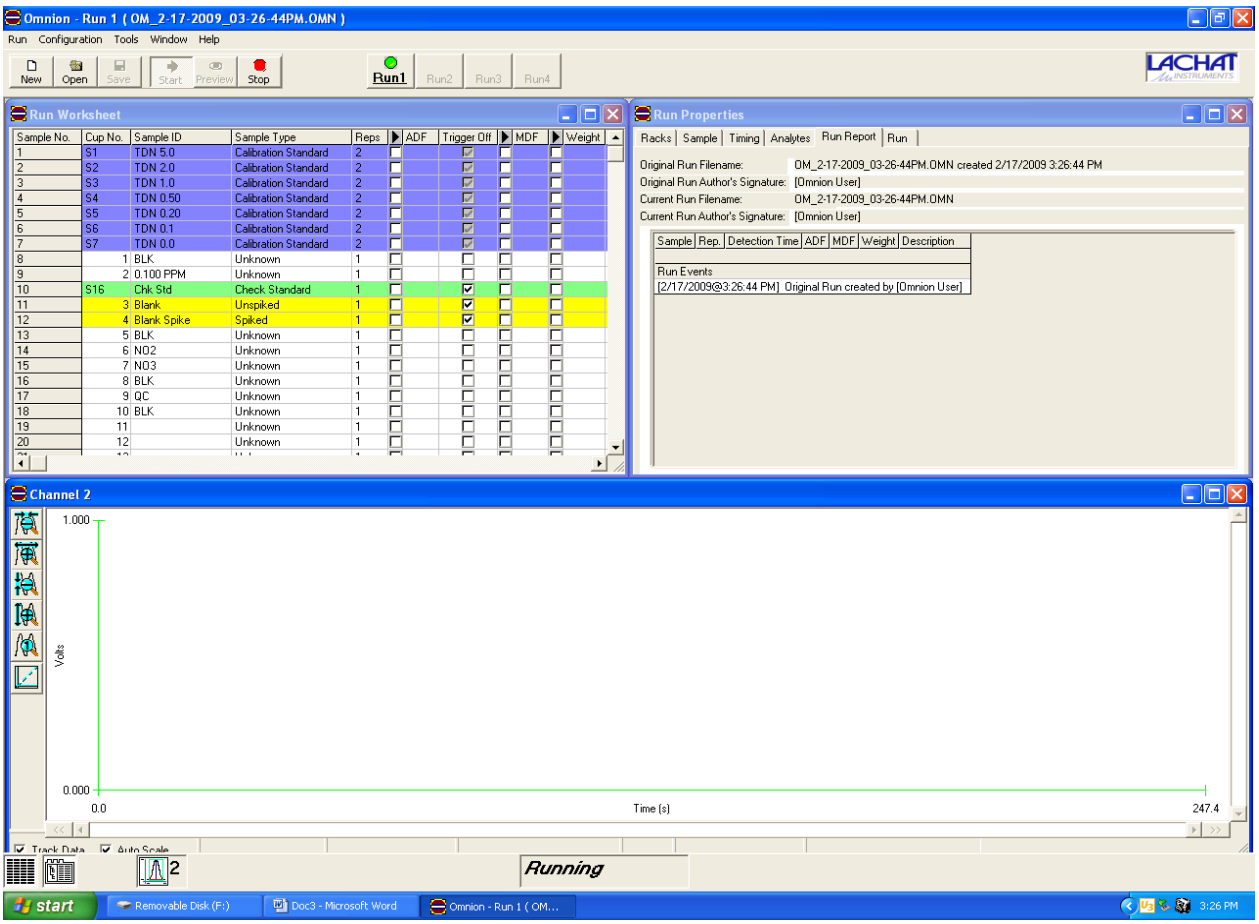
Sample No.	Cup No.	Sample ID	Sample Type	Reps	ADF	Trigger Off	MDF	Weight	Units
1	S1	TDN 5.0	Calibration Standard	2					
2	S2	TDN 2.0	Calibration Standard	2					
3	S3	TDN 1.0	Calibration Standard	2					
4	S4	TDN 0.50	Calibration Standard	2					
5	S5	TDN 0.20	Calibration Standard	2					
6	S6	TDN 0.1	Calibration Standard	2					
7	S7	TDN 0.0	Calibration Standard	2					
8	1	BLK	Unknown	1					
9	2	0.100 PPM	Unknown	1					
10	S16	Chk Std	Check Standard	1					
11	3	Blank	Unspiked	1					
12	4	Blank Spike	Spiked	1					
13	5	BLK	Unknown	1					
14	6	NO2	Unknown	1					
15	7	NO3	Unknown	1					
16	8	BLK	Unknown	1					
17	9	QC	Unknown	1					
18	10	BLK	Unknown	1					
19	11		Unknown	1					
20	12		Unknown	1					
21	13		Unknown	1					
22	14		Unknown	1					
23	15		Unknown	1					
24	16		Unknown	1					
25	17		Unknown	1					
26	18		Unknown	1					
27	19		Unknown	1					
28	20		Duplicate 1	1					
29	21		Duplicate 2	1					
30	21		Unspiked	1					
31	22		Spiked	1					
32	S16	Chk Std	Check Standard	1					
33	23		Unknown	1					
34	24		Unknown	1					
35	25		Unknown	1					
36	26		Unknown	1					
37	27		Unknown	1					
38	28		Unknown	1					
39	29		Duplicate 1	1					
40	30		Duplicate 2	1					
41	30		Unspiked	1					
42	31		Spiked	1					
43	S16	Chk Std	Check Standard	1					
44	32		Unknown	1					
45	33		Unknown	1					
46	34		Unknown	1					
47	35		Unknown	1					
48	36		Unknown	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 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:

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

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

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

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, Maryland Department of Health and Mental Hygiene, *Quality Assurance Plan,* Revision 11, 2011.

APPENDIX A

State of Maryland
DHMH - Laboratories Administration
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		
Samples Analyzed	Within 5 working days		
Calibration Curve	Corr. Coefficient. $\geq$ 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 $\leq$ 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.

_____ Date Reported: _____

Reviewer's Signature & Date

Reagents	ID	Reagents	ID	External QC	
Ammonia Buffer	_____	Oxidizing Reagent	_____	Identification =	_____
Color Reagent	_____	Borate Buffer	_____	True Value =	_____ ppm
				Range =	_____ ppm

State of Maryland
DHMH - Laboratories Administration
DIVISION OF ENVIRONMENTAL CHEMISTRY

STANDARD OPERATING PROCEDURES

Title:	Determination of Ammonia – Low Level Flow Injection Colorimetric Analysis EPA Method 350.1
SOP No.:	IAL-SOP-EPA 3501/R2.0-11
Revision:	2.0 Replaces: 1.0 Effective: 8/18/2011
Laboratory:	Inorganics Analytical Laboratory
POC:	Shahla Ameli amelis@dhmh.state.md.us

Laboratory Supervisor: _____ _____
Signature Date

Division's QA Officer: _____ _____
Signature Date

Division Chief: _____ _____
Signature Date

State of Maryland
DHMH - Laboratories Administration
DIVISION OF ENVIRONMENTAL CHEMISTRY
201 W. Preston Street
Baltimore, MD 21201

REVISION RECORD

Revision	Date	Changes	Made By	Effective Date
0.0	6/3/08	N/A	Shahla Ameli	6/2/08
1.0	12/09	Screen snapshots added to procedure, new Sample Run Log, tracking IDs for standards and reagents	Shahla Ameli	1/10
2.0	8/11	New SOP tracking number and technical and editorial changes	Clair Vares/ Shahla Ameli	8/18/11

TABLE OF CONTENTS

STANDARD OPERATING PROCEDURE

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.

5.1.1.1 XYZ Auto sampler ASX-520 series with sample, standard and dilution racks

5.1.1.2 Manifold or reaction unit

5.1.1.3 Multichannel Reagent Pump RP-100 series

5.1.1.4 Colorimetric Detector

5.1.1.4.1 Flowcell, 10 mm, 80uL, glass flow cell

5.1.1.4.2 630 nm interference filter

5.1.1.5 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 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 50.0 g 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 1 M HCl- Add 83 ml HCl to about 700 mL DI water in a 1 L volumetric flask, and bring up to mark. Use this reagent to rinse the phenol lines that become a brownish color after many runs.

6.1.7 Sodium Hydroxide – EDTA Rinse - In a 1 L volumetric flask, dissolve 65 g sodium hydroxide (NaOH) and 6 g tetrasodium ethylenediamine tetraacetic acid (Na4EDTA) in 800 mL of water. Dilute to the mark after all is dissolved.

6.2 Standards

6.2.1 Ammonia Stock Standard (1000) mg N/L - Dissolve 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:

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

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 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.

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.

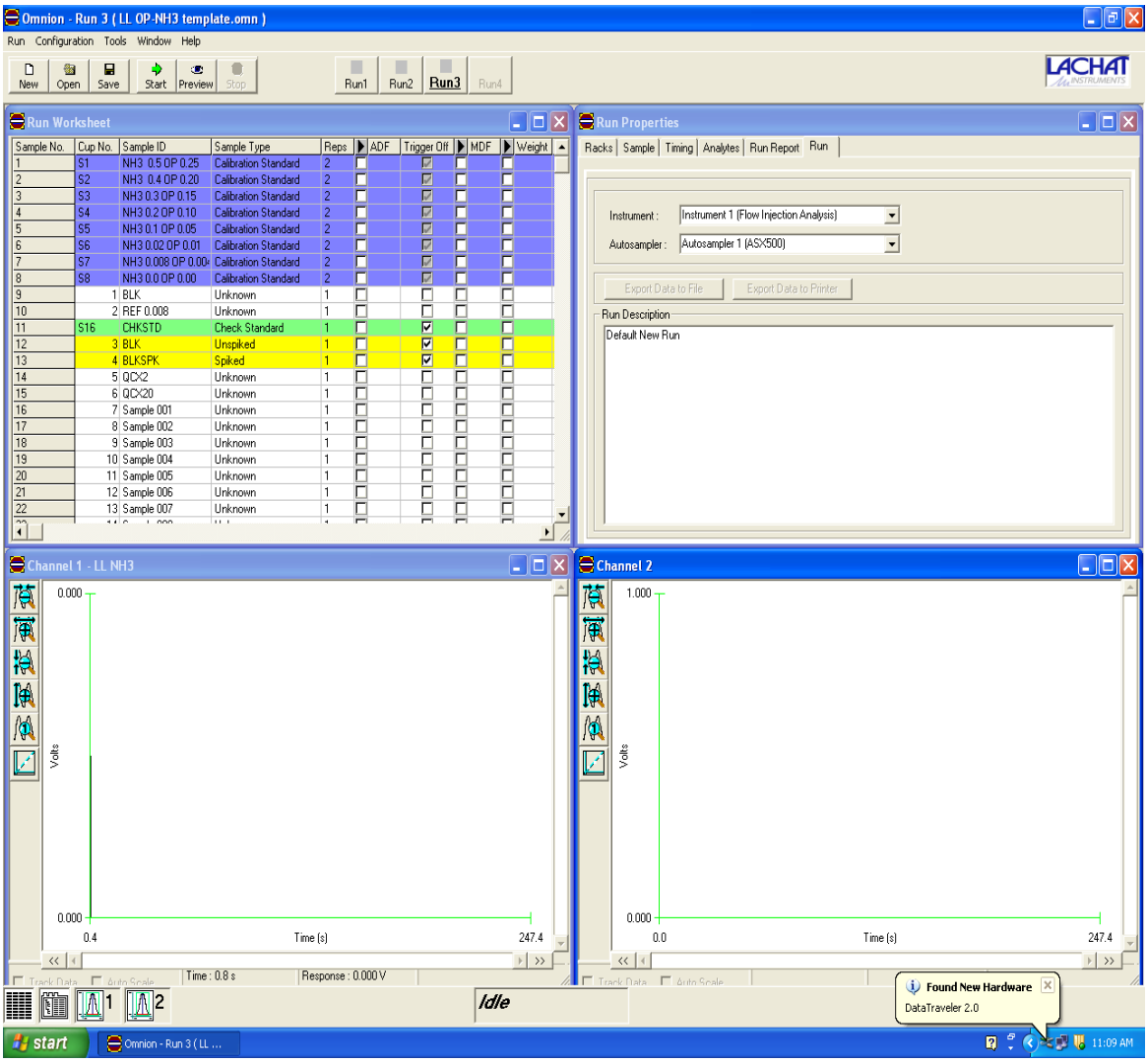
9.1.4 To prevent bubble formation, degas all reagents, except for phenol, with helium for two minutes. Use Helium at 140 kPa (20lb/in2) through a helium degassing tube or a pipette.

9.2 Instrument set-up and sample analysis

9.2.1 Set up manifold as in the diagram.

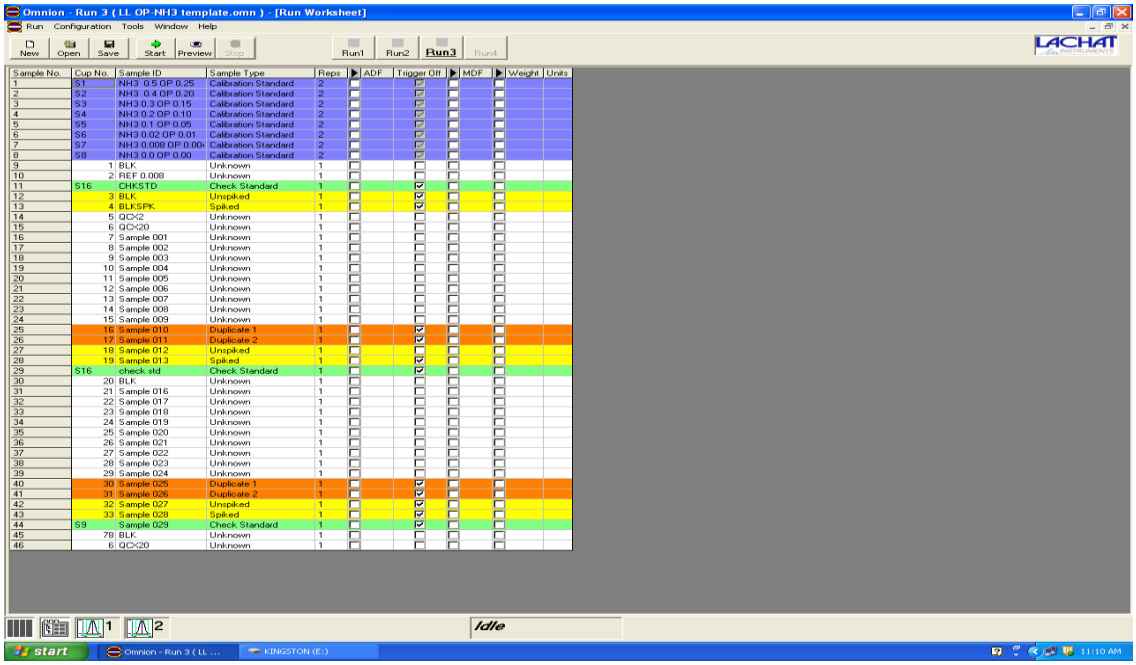
9.2.2 Turn on the Lachat instrument, computer, monitor, and printer.

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.

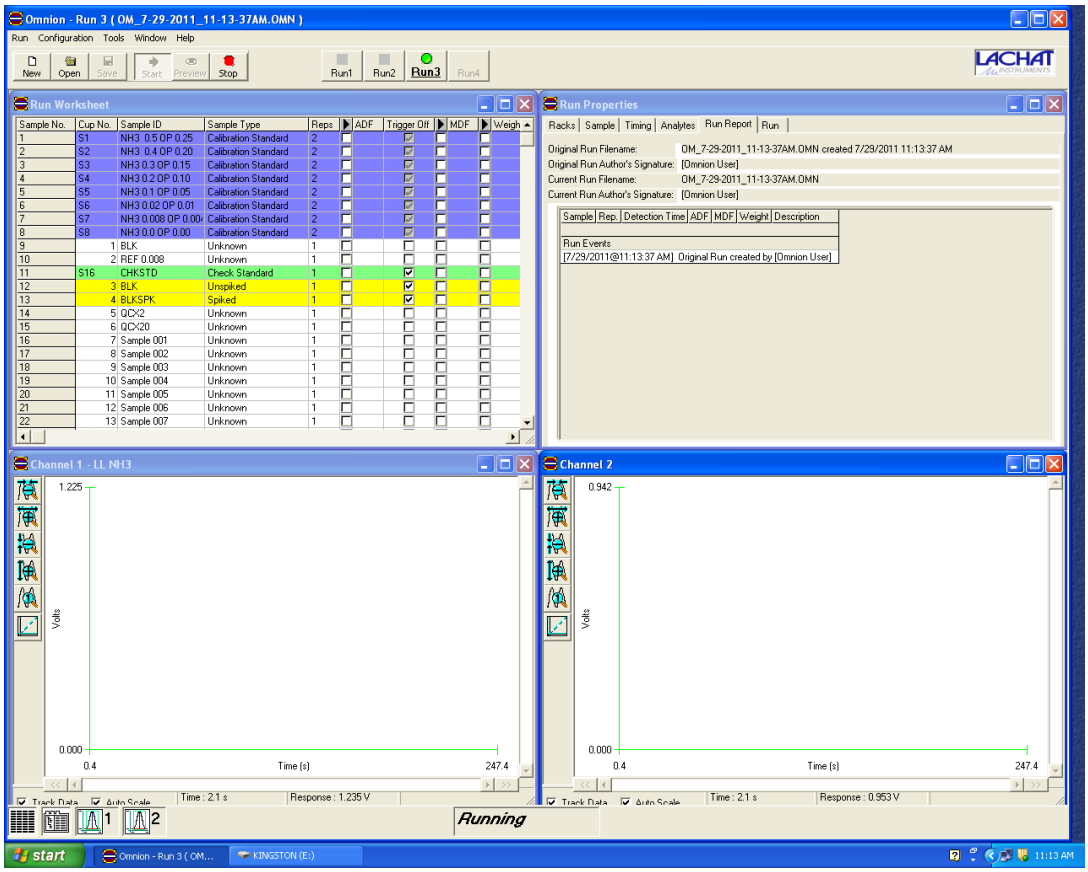


9.3.5 Print a copy of this worksheet by first double clicking on **"Run"** icon and then selecting **"Export Worksheet Data"**.

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 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 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 Report only those results that fall between the lowest and the highest calibration standards. Samples exceeding the highest standard are manually diluted and reanalyzed.

11.2 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, Maryland Department of Health and Mental Hygiene, *Quality Assurance Plan,* Revision 11.0, 2011.

APPENDIX A

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

IAL-SOP-EPA 350.1/R2.0-11

Data Review Checklist

LL Ammonia/EPA Method 350.1

LabNumbers[1]:_____

Date Collected: _____ **Date Analyzed:** _____ **Analyst:** _____

Procedure	Acceptance Criteria	Status (√)	Comments
Holding Time	48 hours @ 4°C 28 days @ –20°C		
Samples Analyzed	Within 5 working days		
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.

_____ Date Reported: _____

Reviewer's Signature & Date

Reagents	ID	Reagents	ID
Sodium Phenolate		Sodium Hypochlorite	
Sodium Nitroprusside		EDTA Buffer	

External QC

Identification = _____

True Value = _____ ppm

Range = _____ ppm

State of Maryland
DHMH - Laboratories Administration
DIVISION OF ENVIRONMENTAL CHEMISTRY

STANDARD OPERATING PROCEDURES

Title:	Determination of Nitrate/Nitrite and Nitrite (Low Level) Flow Injection Colorimetric Analysis EPA Method 353.2
SOP No.:	IAL-SOP-EPA 353.2/R2.0-11
Revision:	2.0 Replaces: 1.0 Effective: 8/18/11
Laboratory:	Inorganics Analytical Laboratory
POC:	Shahla Ameli amelis@dhmh.state.md.us

Laboratory Supervisor: _____ _____
Signature Date

Division's QA Officer: _____ _____
Signature Date

Division Chief: _____ _____
Signature Date

State of Maryland
DHMH - Laboratories Administration
DIVISION OF ENVIRONMENTAL CHEMISTRY
201 W. Preston Street
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REVISION RECORD

Revision	Date	Changes	Made By	Effective Date
0.0	6/3/08	N/A	Shahla Ameli	6/2/08
1.0	12/09	Screen snapshots added to procedure, new Sample Run Log, tracking IDs for standards and reagents	Shahla Ameli	1/10
2.0	8/11	New SOP tracking number, editorial and technical changes	Shahla Ameli	8/18/11

TABLE OF CONTENTS

STANDARD OPERATING PROCEDURES

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

4.3 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 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 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 (Na2EDTA'2H2O) 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 weekly.

6.1.2 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.

6.1.3 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 Ricca Chemicals with expiration date; cat. No. 5459-4. 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 Ricca Chemicals with expiration date; cat. No. 5461-4. 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^oC. 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. Adust 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 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.

8.8 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.

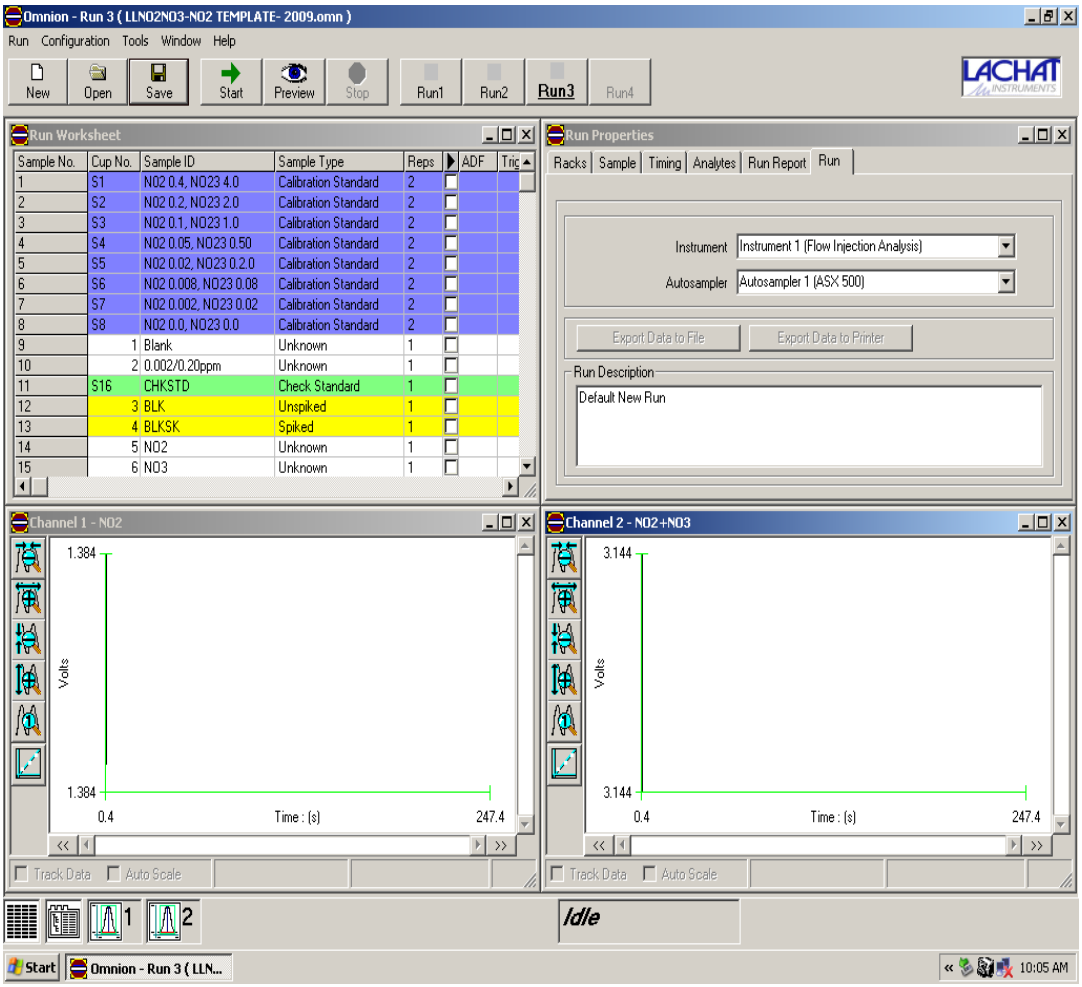
9.1.3 To prevent bubble formation, degas all reagents with helium, except those specified not to. Use helium at 140 Pa (20lb/in2)

9.2 Instrument calibration and sample analysis

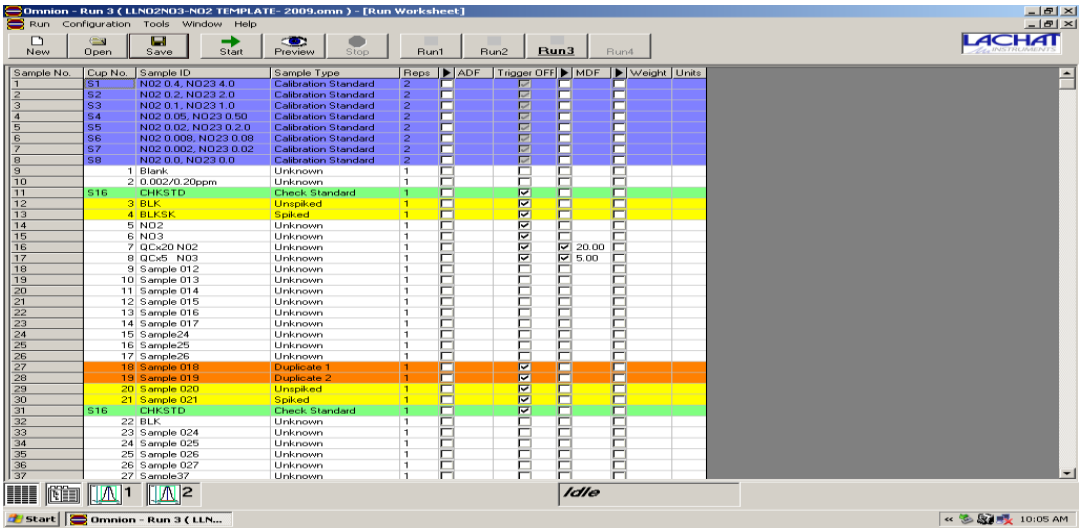
9.2.1 Set up manifold as in the method's manifold diagram.

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.



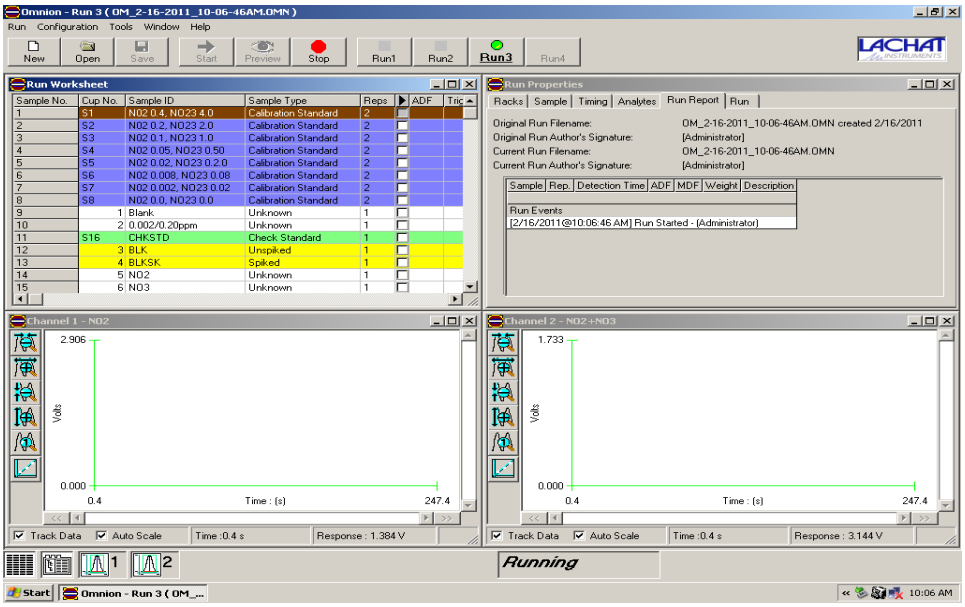
9.2.5 Enter all the sample, sample spike, QC and standard information in the run worksheet and print out the list.

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 The reduction efficiency of the cadmium coil is calculated as followings:

$$\frac{NO_3 \text{ (peak height)}}{NO_2 \text{ (peak height)}} \text{ X } 100$$

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

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

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

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

11.0 WASTE MANAGEMENT

11.1 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.

12.0 REFERENCES

12.1 EPA Method 353.2, *Methods for the Determination of Inorganic Substances in Environmental Samples,* Revision 2.0*,* August 1993.

12.2 American Public Health Association, *Standard Methods for the Examination of Water and Wastewater, 21st Edition, p. 4-125, Method 4500- NO_3^-, 2005*

12.3 Lachat Instruments, Methods Manual for the *Quikchem Automated Ion Analyzer*, Method 10-107-04-1-A.

12.4 Division of Environmental Chemistry, Maryland Department of Health and Mental Hygiene, *Quality Assurance Plan,* Revision 10.0, 2011.

Appendix A

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

Data Review Checklist

LL Nitrate + Nitrite/EPA Method 353.2

Lab Numbers[1]: _____

Date Collected: _____ Date Analyzed: _____ Analyst: _____

Procedure	Acceptance Criteria	Status (√)	Comments
Holding Time	48 hours @ 4°C 28 days @ –20°C		
Samples Analyzed	Within 5 working days		
Calibration Curve	Corr. Coefficient. ≥ 0.9950		
Reagent Blank	< 0.02 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		
	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	1		
Measured Values	Within calibration range (0.02 – 4.0 ppm)		
Diluted Samples	Correct final calculations		
Changes/Notes	Clearly stated		

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

_____ Date Reported: _____

Reviewer's Signature & Date

Reagents ID

Ammonia Buffer _____

Color Reagent _____

External QC

Identification = _____

True Value = _____ ppm

Range = _____ ppm

State of Maryland
DHMH - Laboratories Administration
DIVISION OF ENVIRONMENTAL CHEMISTRY

STANDARD OPERATING PROCEDURES

Title:	Determination of Orthophosphate- Low Level Flow Injection Colorimetric Analysis EPA Method 365.1
SOP No.:	IAL-SOP-EPA 365.1/R2.0-11
Revision:	2.0 Replaces: 1.0 Effective: 8/18/2011
Laboratory:	Inorganics Analytical Laboratory
POC:	Shahla Ameli amelis@dhmh.state.md.us

Laboratory Supervisor: _____ _____
Signature Date

Division's QA Officer: _____ _____
Signature Date

Division Chief: _____ _____
Signature Date

State of Maryland
DHMH - Laboratories Administration
DIVISION OF ENVIRONMENTAL CHEMISTRY
201 W. Preston Street
Baltimore, MD 21201

REVISION RECORD

Revision	Date	Changes	Made By	Effective Date
0.0	6/3/08	N/A	Shahla Ameli	6/2/08
1.0	12/09	Screen snapshots added to procedure, new Sample Run Log, tracking IDs for standards and reagents	Shahla Ameli	1/10
2.0	8/11	New SOP tracking number, technical and editorial changes	Shahla Ameli	8/18/11

TABLE OF CONTENTS

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

5.1.1.2 Manifold or reaction unit

5.1.1.3 Multichannel Reagent Pump RP-100 series

5.1.1.4 Colorimetric Detector

5.1.1.4.1 Flowcell, 10 mm, 80uL, glass flow cell

5.1.1.4.2 880 nm interference filter

5.1.1.5 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) - Dissolve 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 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.

8.7 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.

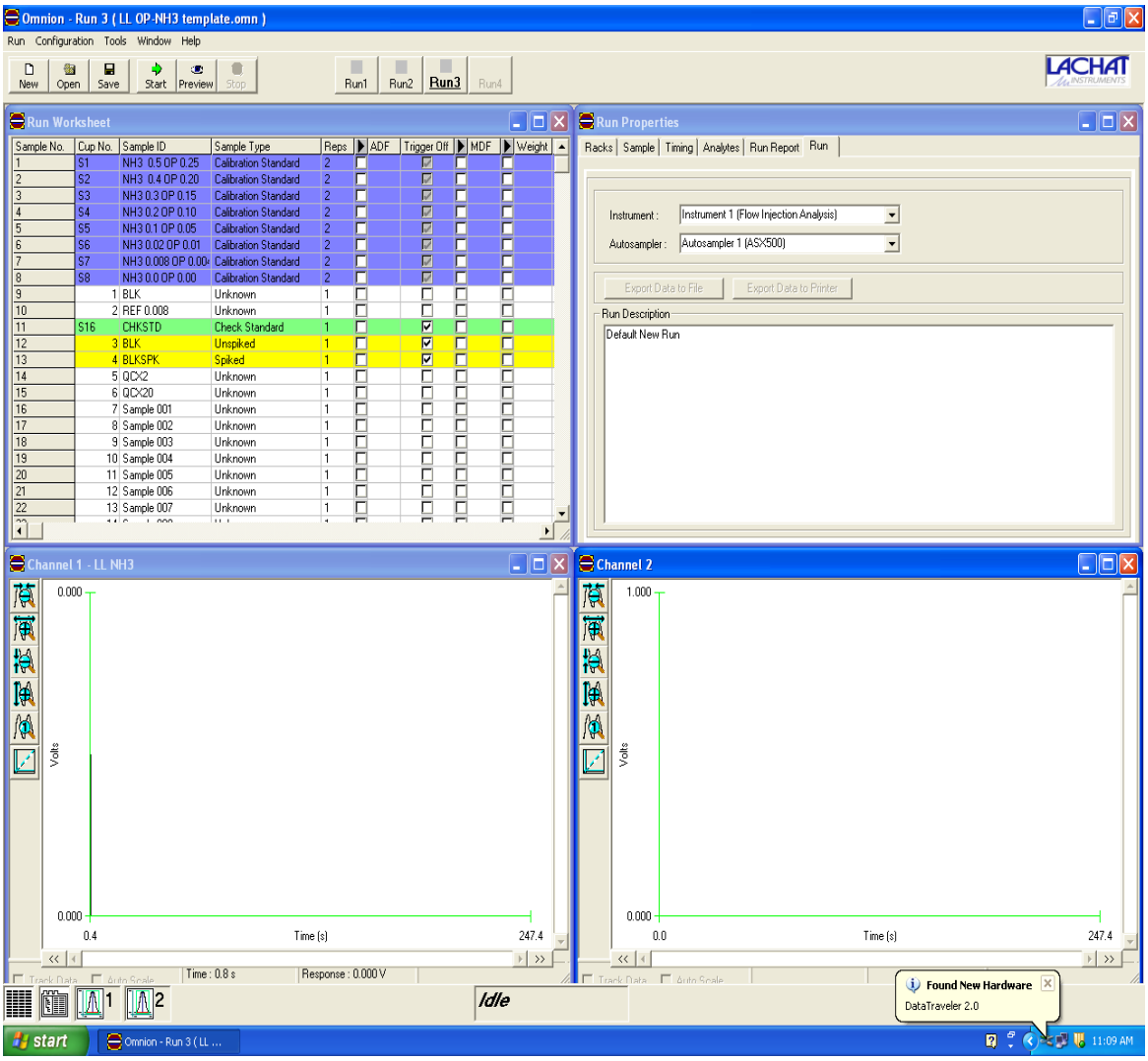
9.1.2 To prevent bubble formation, degas all reagents, except those specified by the method with helium. Use He at 140 kPa (20lb/in2) through a helium degassing tube or a pipette for two minutes.

9.2 Instrument set-up and sample analysis

9.2.1 Set up manifold as in the diagram.

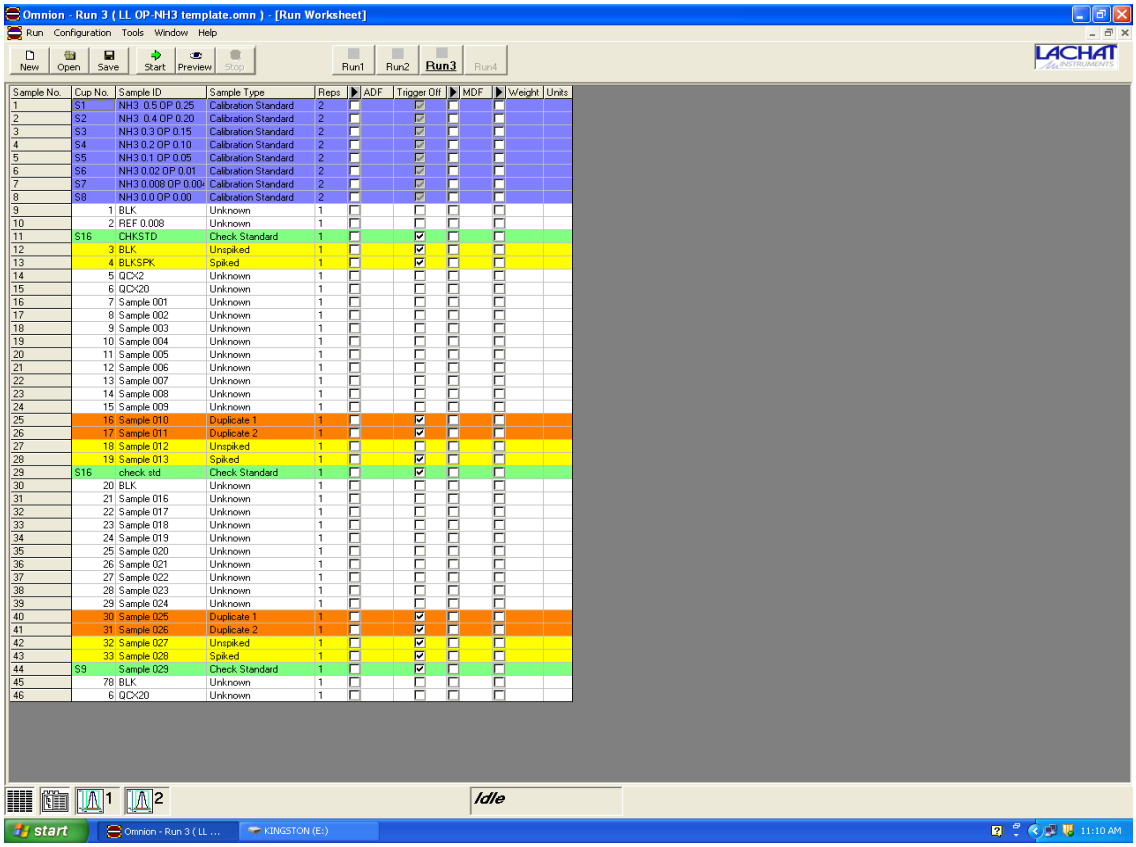
9.2.2 Turn on the Lachat instrument, computer, monitor, and printer.

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.



Sample No.	Cup No.	Sample ID	Sample Type	Reps	ADF	Trigger Off	MDF	Weight	Units
1	S1	NH3 0.5 OP 0.25	Calibration Standard	2					
2	S2	NH3 0.4 OP 0.20	Calibration Standard	2					
3	S3	NH3 0.3 OP 0.15	Calibration Standard	2					
4	S4	NH3 0.2 OP 0.10	Calibration Standard	2					
5	S5	NH3 0.1 OP 0.05	Calibration Standard	2					
6	S6	NH3 0.02 OP 0.01	Calibration Standard	2					
7	S7	NH3 0.008 OP 0.00	Calibration Standard	2					
8	S8	NH3 0.0 OP 0.00	Calibration Standard	2					
9	1	BLK	Unknown	1					
10	2	REF 0.008	Unknown	1					
11	S16	CHKSTD	Check Standard	1					
12	3	BLK	Unspiked	1					
13	4	BLKSPK	Spiked	1					
14	5	QCX2	Unknown	1					
15	6	QCX20	Unknown	1					
16	7	Sample 001	Unknown	1					
17	8	Sample 002	Unknown	1					
18	9	Sample 003	Unknown	1					
19	10	Sample 004	Unknown	1					
20	11	Sample 005	Unknown	1					
21	12	Sample 006	Unknown	1					
22	13	Sample 007	Unknown	1					
23	14	Sample 008	Unknown	1					
24	15	Sample 009	Unknown	1					
25	16	Sample 010	Duplicate 1	1					
26	17	Sample 011	Duplicate 2	1					
27	18	Sample 012	Unspiked	1					
28	19	Sample 013	Spiked	1					
29	S16	check std	Check Standard	1					
30	20	BLK	Unknown	1					
31	21	Sample 016	Unknown	1					
32	22	Sample 017	Unknown	1					
33	23	Sample 018	Unknown	1					
34	24	Sample 019	Unknown	1					
35	25	Sample 020	Unknown	1					
36	26	Sample 021	Unknown	1					
37	27	Sample 022	Unknown	1					
38	28	Sample 023	Unknown	1					
39	29	Sample 024	Unknown	1					
40	30	Sample 025	Duplicate 1	1					
41	31	Sample 026	Duplicate 2	1					
42	32	Sample 027	Unspiked	1					
43	33	Sample 028	Spiked	1					
44	S9	Sample 029	Check Standard	1					
45	78	BLK	Unknown	1					
46	6	QCX20	Unknown	1					

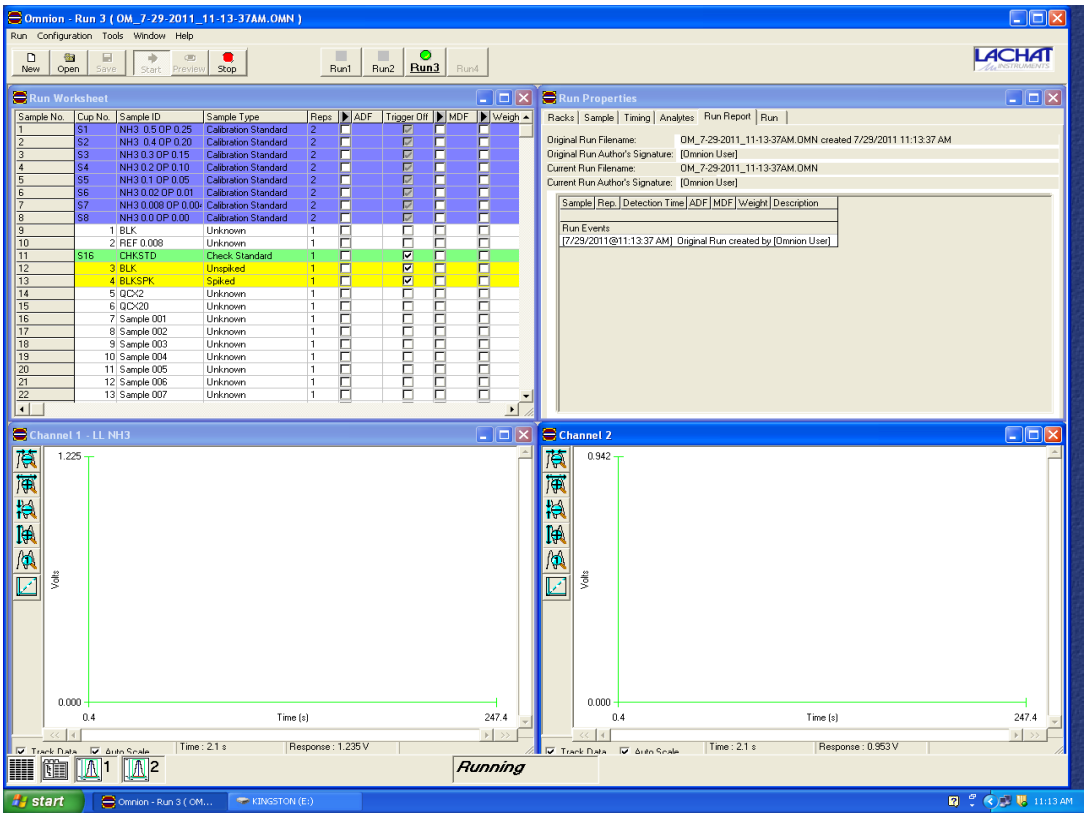
9.3.5 Print a copy of this worksheet by first double clicking on **"Run"** icon and then selecting **"Export Worksheet Data"**.

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 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

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.

9.2.12 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 Report only those results that fall between the lowest and the highest calibration standards. Samples exceeding the highest standard are manually diluted and reanalyzed.

11.2 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

12.1 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. For more information consult the “Waste Management

12.3 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, Maryland Department of Health and Mental Hygiene, *Quality Assurance Plan,* Revision 11.0, 2011.

APPENDIX A

State of Maryland
DHMH - Laboratories Administration
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		
Samples Analyzed	Within 5 working days		
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.

_____ Date Reported: _____
Reviewer's Signature & Date

Reagents	ID
Color Reagent	_____
Ascorbic Acid	_____
	_____

External QC
Identification = _____
True Value = _____ ppm
Range = _____ ppm

APPENDIX B

State of Maryland
DHMH - Laboratories Administration
DIVISION OF ENVIRONMENTAL CHEMISTRY
Inorganics Analytical Laboratory

Data Review Checklist

LL Orthophosphate/EPA Method 365.1
LL Ammonia/ Lachat EPA Method 350.1

Lab Numbers[1]: _____

Date Collected: _____ **Date Analyzed:** _____ **Analyst:** _____

Procedure	Acceptance Criteria	Status (√)	Comments
Holding Time	48 hours @ 4°C; 28 days @ –20°C		
Samples Analyzed	Within 5 working days		
Calibration Curve	Corr. Coefficient. ≥ 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 run		
	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.

_____ Date Reported: _____
Reviewer's Signature & Date

Reagents	ID	Reagents	ID
Sodium Phenolate		Color Reagent	
Sodium Nitroprusside		Ascorbic Acid	
Sodium Hypochlorite			
EDTA Buffer			

External QC
Identification = _____
True Value = _____ ppm
Range = _____ ppm

State of Maryland
DHMH - Laboratories Administration
DIVISION OF ENVIRONMENTAL CHEMISTRY

STANDARD OPERATING PROCEDURES

Title:	Determination of Total Dissolved Phosphorus Flow Injection Colorimetric Analysis EPA Method 365.1
SOP No.:	IAL-SOP-EPA 365.1/R2.0-11
Revision:	2.0 Replaces: 1.0 Effective: 8/18/2011
Laboratory:	Inorganics Analytical Laboratory
POC:	Shahla Ameli amelis@dhmh.state.md.us

Laboratory Supervisor: _____ _____
Signature Date

Division's QA Officer: _____ _____
Signature Date

Division Chief: _____ _____
Signature Date

State of Maryland
DHMH - Laboratories Administration
DIVISION OF ENVIRONMENTAL CHEMISTRY
201 W. Preston Street
Baltimore, MD 21201

REVISION RECORD

Revision	Date	Changes	Made By	Effective Date
0.0	6/3/08	N/A	Shahla Ameli	6/2/08
1.0	12/09	Screen snapshots added to procedure, new Sample Run Log, tracking IDs for standards and reagents	Shahla Ameli	1/10
2.0	1/11	New SOP tracking number, technical and editorial changes	J Freeman-Scott, Shahla Ameli	8/18/2011

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STANDARD OPERATING PROCEDURES

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

6.2.4 Spiking Solution - Use the mixed intermediate standard (6.2.2) as

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 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 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 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

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

9.2.1 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.

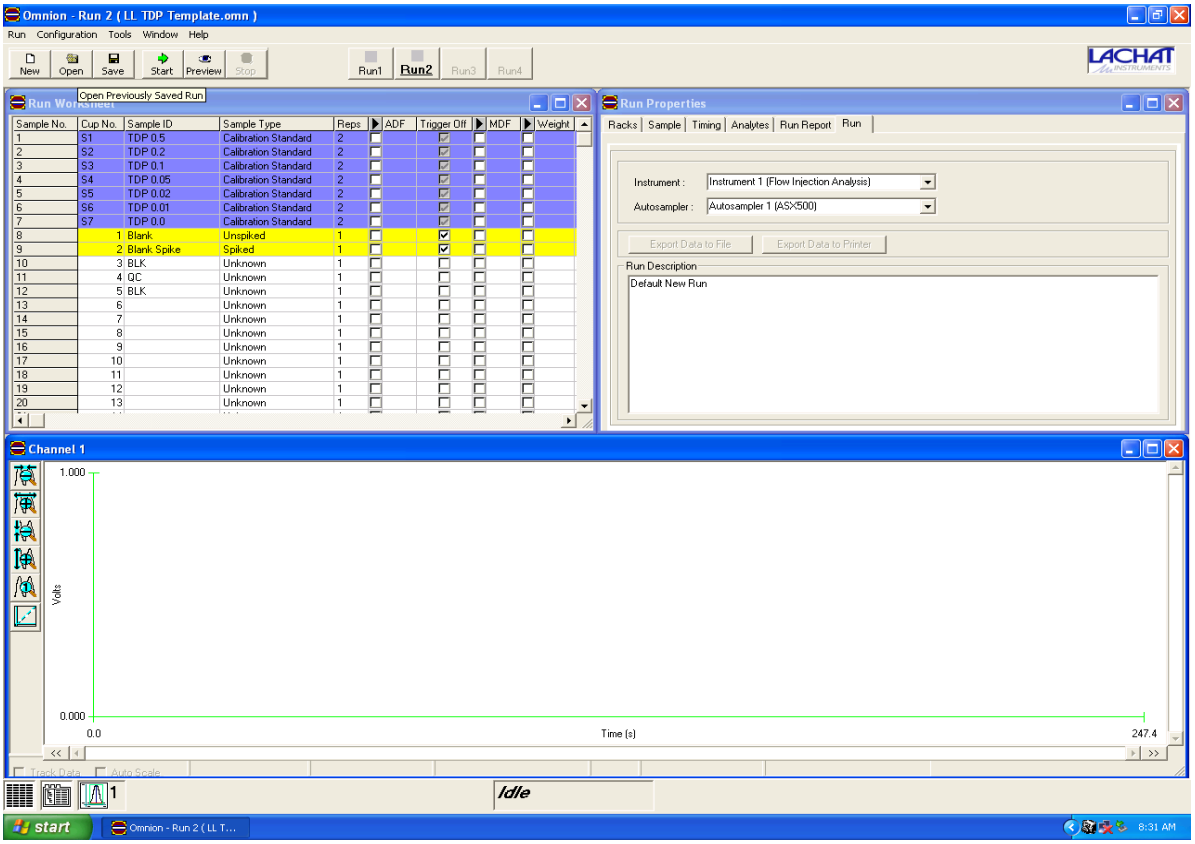
9.2.7 Get the data reviewed by a designated scientist, and then, report the results on the Analysis Request Forms.

9.3 Instrument set-up and sample analysis

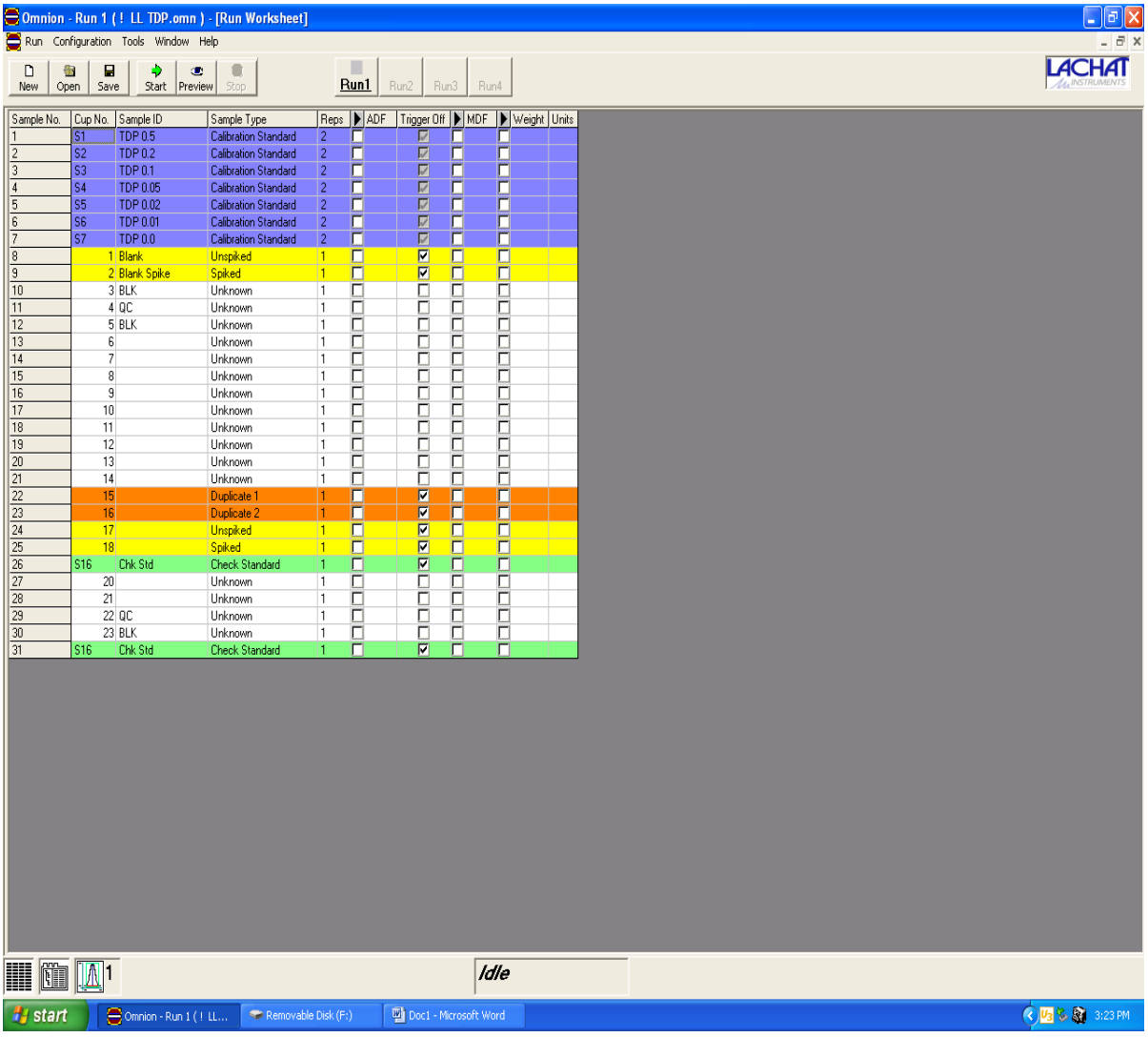
9.3.1 Set up manifold as in the diagram.

9.3.2 Turn on the Lachat instrument, computer, monitor, and printer.

9.3.3 Double click on 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	☐	☑	☐		

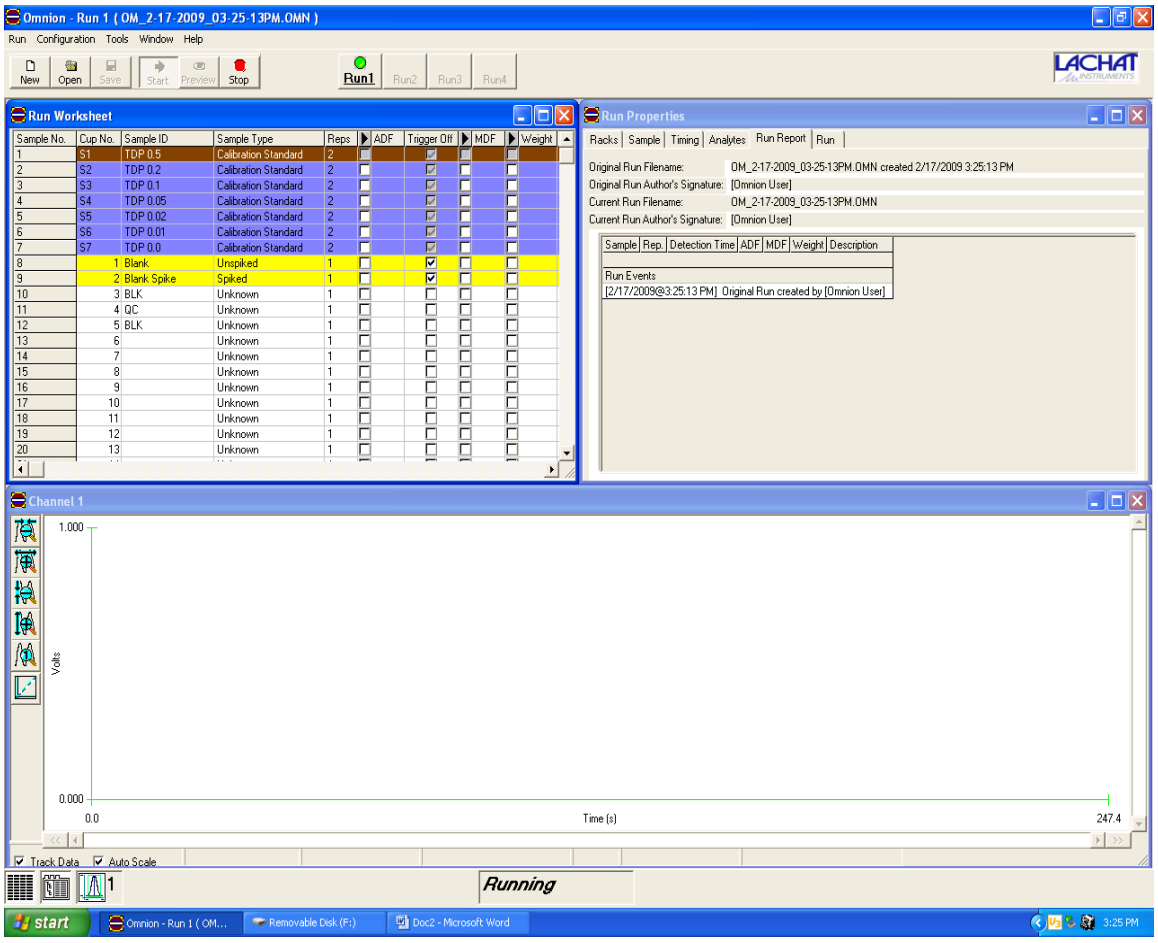
9.3.5 Print a copy of this worksheet by first double clicking on **"Run"** icon and then selecting **"Export Worksheet Data"**.

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.

9.2.12 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 (6.1.5) 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:

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

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

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

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 Division of Environmental Chemistry, Maryland Department of Health and Mental Hygiene, *Quality Assurance Plan,* Revision 11.0, 2011.

APPENDIX A

State of Maryland
DHMH - Laboratories Administration
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 Collected: _____ **Date Digested:** _____ **Date Analyzed:** _____

Procedure	Acceptance Criteria	Status (√)	Comments
Holding Time	48 hours @ 4°C 28 days @ –20°C		
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.

_____ Date Reported: _____
Reviewer's Signature & Date

	ID	Reagents	ID		External QC
Reagents					
Color Reagent	_____	Oxidizing Reagent	_____	Identification =	_____
Ascorbic Acid	_____	Borate Buffer	_____	True Value =	_____ ppm
1M HCl	_____			Range =	_____ ppm

State of Maryland
DHMH - Laboratories Administration
DIVISION OF ENVIRONMENTAL CHEMISTRY

STANDARD OPERATING PROCEDURES

Title:	Determination of Total Organic Carbon (Standard Method 5310 B)
SOP No.:	IAL-SOP-SM 5310 B/R1.0-11
Revision:	1.0 Replaces: 0.0 Effective: 8/18/11
Laboratory:	Inorganics Analytical Laboratory
POC:	Reza Hajarian HajarianR@dhmh.state.md.us

Laboratory Supervisor: _____ _____
Signature Date

Division's QA Officer: _____ _____
Signature Date

Division Chief: _____ _____
Signature Date

State of Maryland
DHMH - Laboratories Administration
DIVISION OF ENVIRONMENTAL CHEMISTRY
201 W. Preston Street
Baltimore, MD 21201

REVISION RECORD

Revision	Date	Changes	Made By	Effective Date
0.0	12/09	N/A	Taiyin Wei	1/10
1.0	8/11	New procedure section, new SOP tracking number	Reza Hajarian	1.0

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STANDARD OPERATING PROCEDURES

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 ^{o}C. 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 with 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 1 liter volumetric flasks respectively and mix thoroughly. Transfer to a reagent bottle, 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 repectively. 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 two vials filled with blank (0 ppm) and 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

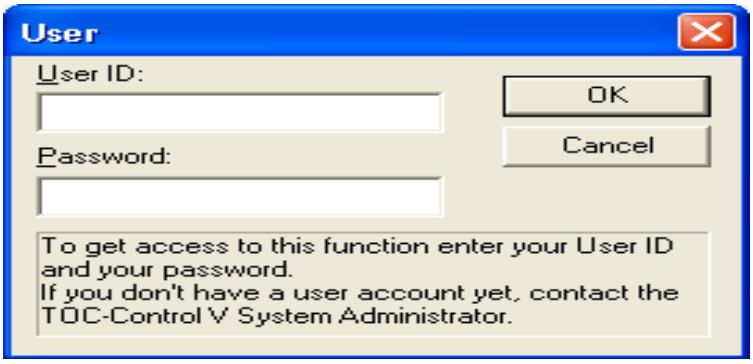
9.1 Analysis Flow

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

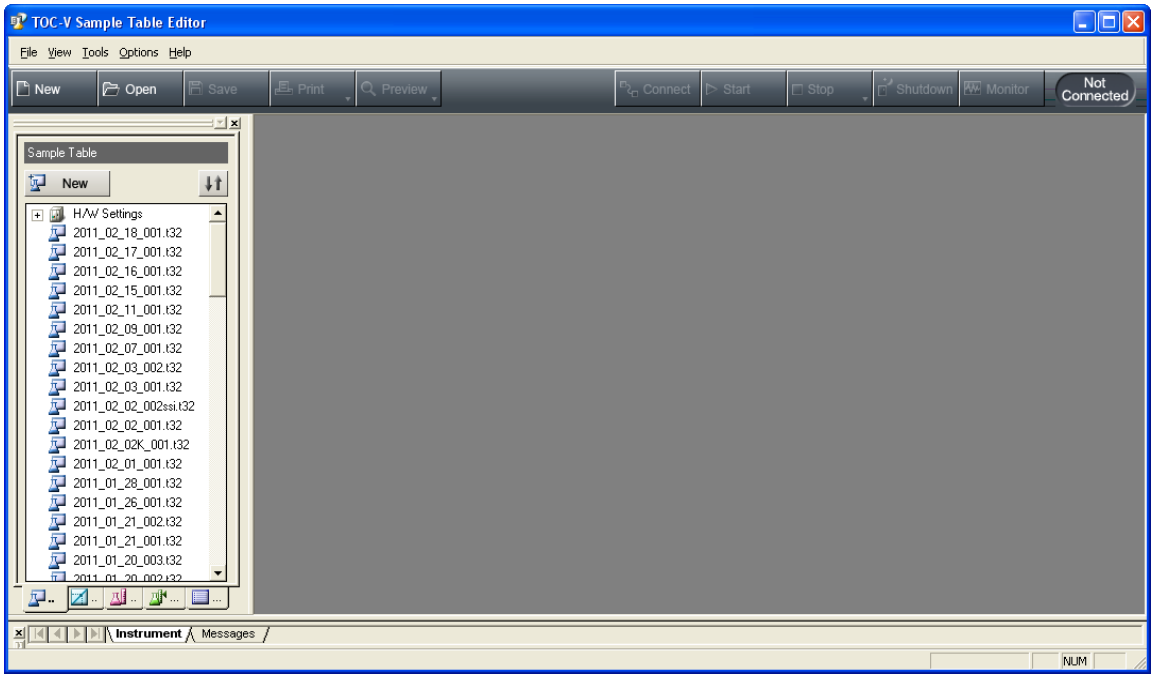
9.1.2 Switch on TOC-V_{cph} Analyzer, which undergoes the initialization sequence.

9.1.3 Turn on the computer.

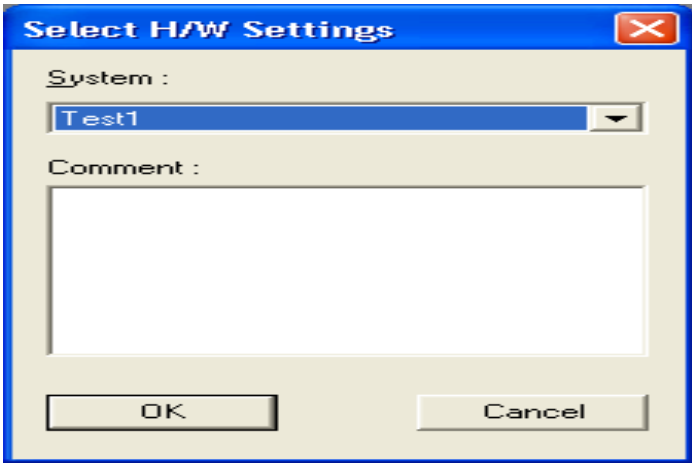
9.1.4 Double click on TOC-V Sample Table Editor icon on the Monitor. The “User” window is displayed.



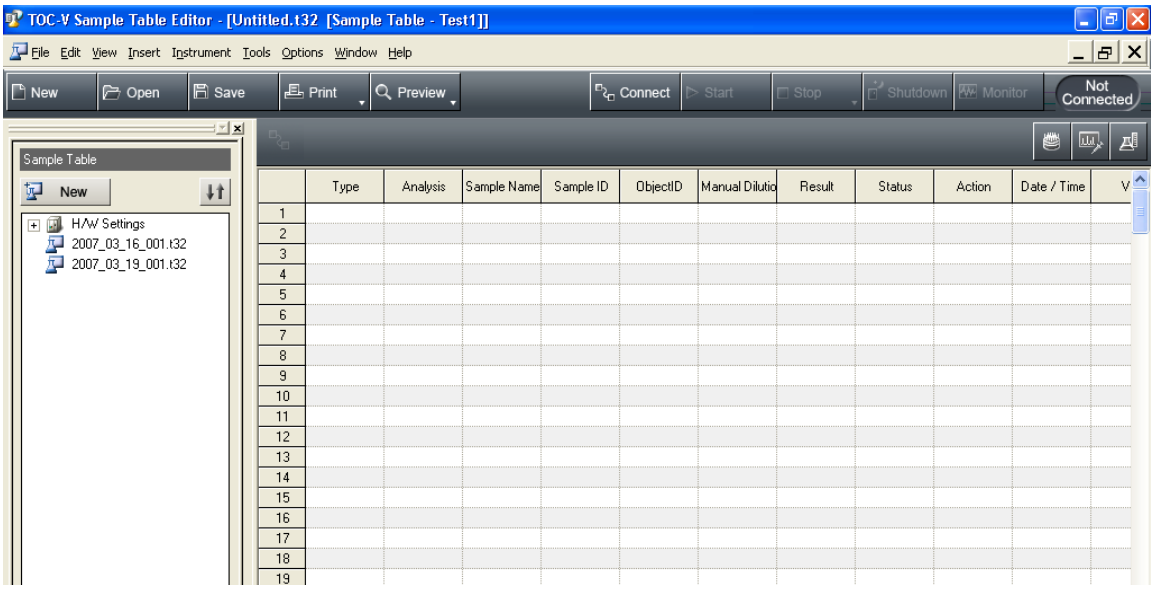
9.1.5 Click OK on the User. A TOC-V Sample Table Editor is opened.



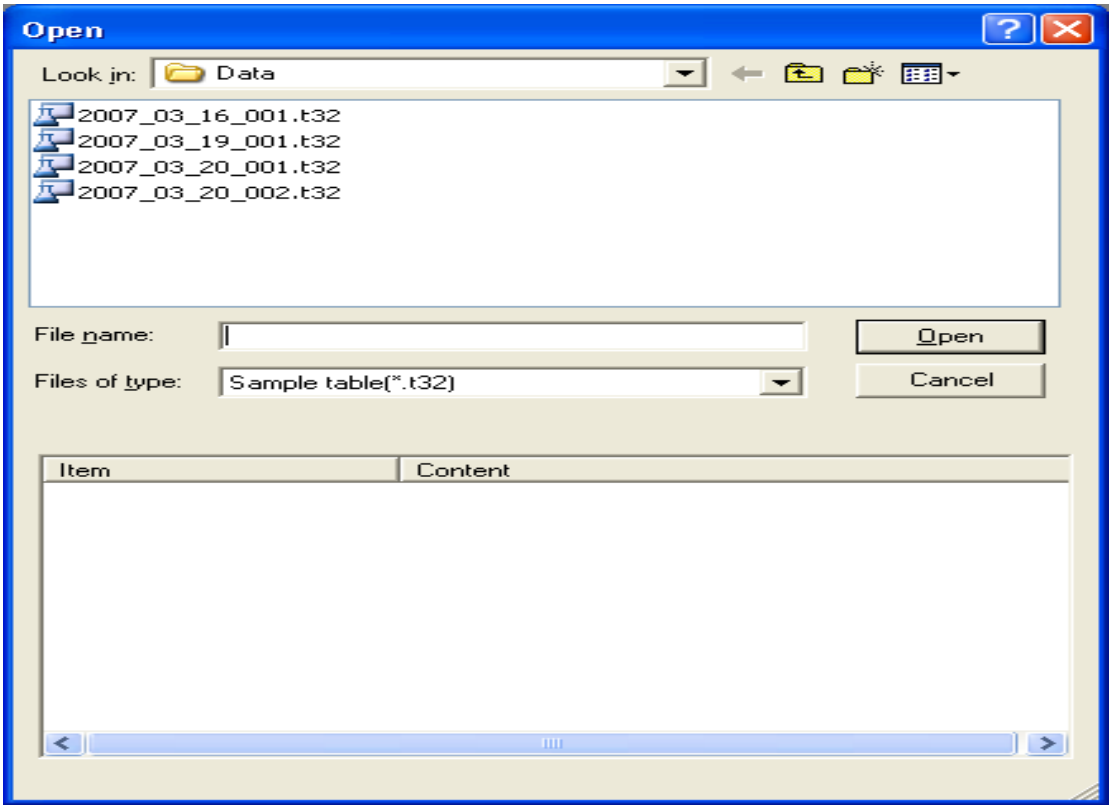
9.1.6 Click “New” on Sample Table Editor, The”Select H/W Settings” window is displayed.



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

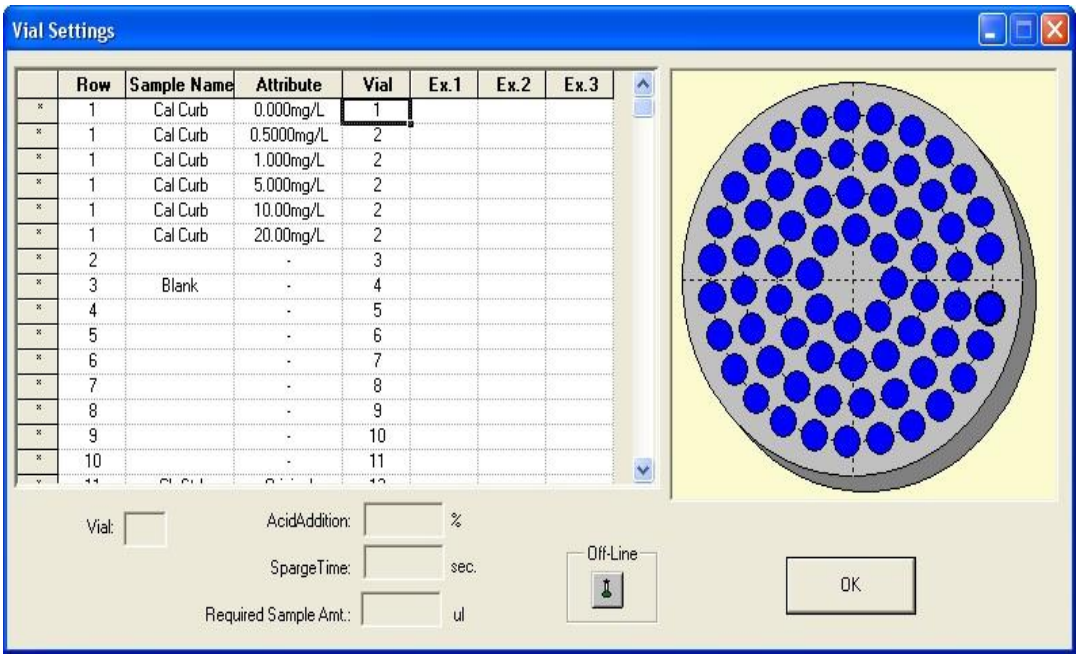


9.1.8 On TOC-V Sample Table Editor, select File, Import Schedule File in the menu Bar. The Open dialog is displayed.



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

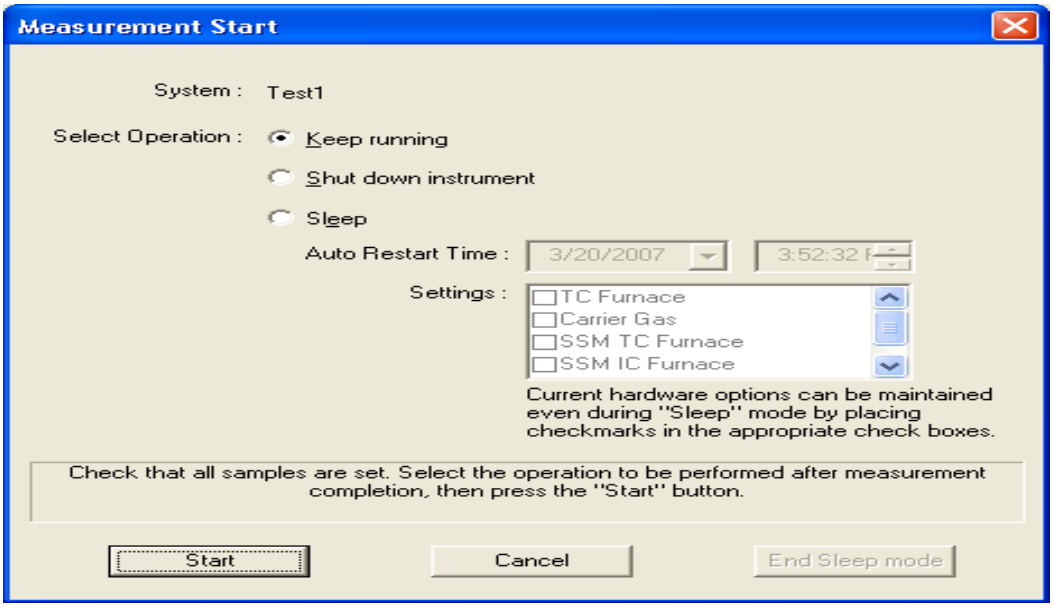
blank, and a check standard are analyzed after every 10 samples. A blank, a check standard and a QC should also be included at the end of each run.

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.

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 10.0, 2010.

APPENDICES

Appendix A – Data Review Checklist
Appendix B – TOC Run Log

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 ^{o}C for filtered samples; 28 days @ 4 ^{o}C for samples acidified to pH < 2 with HCl		
Samples Analyzed	Within 5 working days		
Calibration Curve	Corr. Coeff. $\geq$ 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 $\leq$ 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.

_____ Date Reported: _____
Signature & Date

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

[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

State of Maryland
DHMH - Laboratories Administration
DIVISION OF ENVIRONMENTAL CHEMISTRY

STANDARD OPERATING PROCEDURES

Title:	Determination of Total Suspended Solids (Standard Method 2540 D)
SOP No.:	IAL-SOP-SM 2540 D/R1.1-11
Revision:	1.1 Replaces: 1.0 Effective: 8/18/11
Laboratory:	Inorganics Analytical Laboratory
POC:	Reza Hajarian HajarianR@dhmh.state.md.us

Laboratory Supervisor: _____ _____
Signature Date

Division's QA Officer: _____ _____
Signature Date

Division Chief: _____ _____
Signature Date

State of Maryland
DHMH - Laboratories Administration
DIVISION OF ENVIRONMENTAL CHEMISTRY
201 W. Preston Street
Baltimore, MD 21201

REVISION RECORD

Revision	Date	Changes	Made By	Effective Date
0.0	12/09	N/A	Taiyin Wei	1/10
1.0	1/11	New SOP tracking number	Asoka Katumuluwa	
1.1	8/11	Updated procedure to reflect Proweigh filters	Moses Obura, S. Ameli	8/18/11

TABLE OF CONTENTS

STANDARD OPERATING PROCEDURES

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 2.0 µm (or smaller) nominal pore size.

1.3 This method is suitable for the determination of solids in portable, 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.

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

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

5.1.8 Filter Assembly for in house prepared pre-wash and pre-weigh filters

5.1.8.1 Filters – Glass microfiber, 24 mm diameter, 1.5 µm retention, cat # 934-AH, Whatman

5.1.8.2 Flask – Filtering with tabulation, 1000 ml, cat # 10-180F, Fisher

5.1.9 Filter Assembly for using commercial 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 the 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 PROCEDURE

9.7 Prepare a sample run log starting with a deionized water blank, replicates for every tenth samples or one per batch, and a QC when needed using either a log sheet (Appendix B) or on StarLIMS.

9.8 Preparation of filters (skip this step if ProWeigh filters are to be used)

9.3 Sample Analysis

9.3.1 Turn on the computer. Click on the "**LabX direct balance**" icon.

9.3.2 Click to open TSS folder, select TSS template and enter the sample list. Click file and save as and enter the new file name as "**yy-mo-day**".

9.3.3 Check the balance with minimum three weights and record in the log book.

9.3.4 Prepare your 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).

9.3.5 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.

9.3.6 Using forceps, carefully place filter in the filter vacuum assembly. Squirt some distilled water in the filter to wet then turn on the vacuum pump.

9.3.7 Dispense measured volume in the corresponding filter as per your run log.

9.3.8 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.

9.3.9 After all liquid has passed through the filter, disconnect the suction flask from the pump tubing first to release the pressure. Then turn off the vacuum. Remove filter and place it back in the original metal pan.

9.3.10 Dry at 103 ^{o} to 105 ^{o}C overnight, cool in a desiccator for 2 hours and determine the 1st final weight.

9.3.11 Return the filters into the oven for at least one hour, cool in desiccators for two hours, and determine the 2nd final weight.

9.3.12 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.

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:

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

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

13.1 United States Environmental Protection Agency, *Methods for Chemical Analysis of Water and Waste,* Method Number 160.2, August, 1993

13.2 American Public Health Association, *Standard Methods for the Examination of Water and Wastewater,* Method Number 2540 D, 21st Edition, 2005

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

APPENDICES

Appendix A – Data Review Checklist
Appendix B – Sample Run Log

APPENDIX A

State of Maryland
DHMH - Laboratories Administration
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.

_____ Date Reported: _____
Reviewer's Signature & Date

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

[2]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 – 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)	TDS (mg/L)	Average & RPD

State of Maryland
DHMH - Laboratories Administration
DIVISION OF ENVIRONMENTAL CHEMISTRY

STANDARD OPERATING PROCEDURES

Title:	Determination of Particulate Phosphorus Flow Injection Colorimetric Analysis EPA Method 365.1
SOP No.:	IAL-SOP-EPA 365.1/R2.0-11
Revision:	2.0 Replaces: 1.0 Effective: 8/18/2011
Laboratory:	Inorganics Analytical Laboratory
POC:	Shahla Ameli amelis@dhmh.state.md.us

Laboratory Supervisor: _____ _____
Signature Date

Division's QA Officer: _____ _____
Signature Date

Division Chief: _____ _____
Signature Date

State of Maryland
DHMH – Laboratories Administration
DIVISION OF ENVIRONMENTAL CHEMISTRY
201 W. Preston Street
Baltimore, MD 21201

REVISION RECORD

Revision	Date	Changes	Made By	Effective Date
0.0	6/3/08	N/A	Shahla Ameli	6/2/08
1.0	12/09	Screen snapshots added to procedure, new Sample Run Log, tracking IDs for standards and reagents	Shahla Ameli	1/10
2.0	8/7/11	New SOP tracking number, editorial and technical changes	Shahla Ameli	8/18/11

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STANDARD OPERATING PROCEDURES

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.

2.2 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:

5.1.1.1 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 tetraacetic acid (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^{o}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

7.1 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.

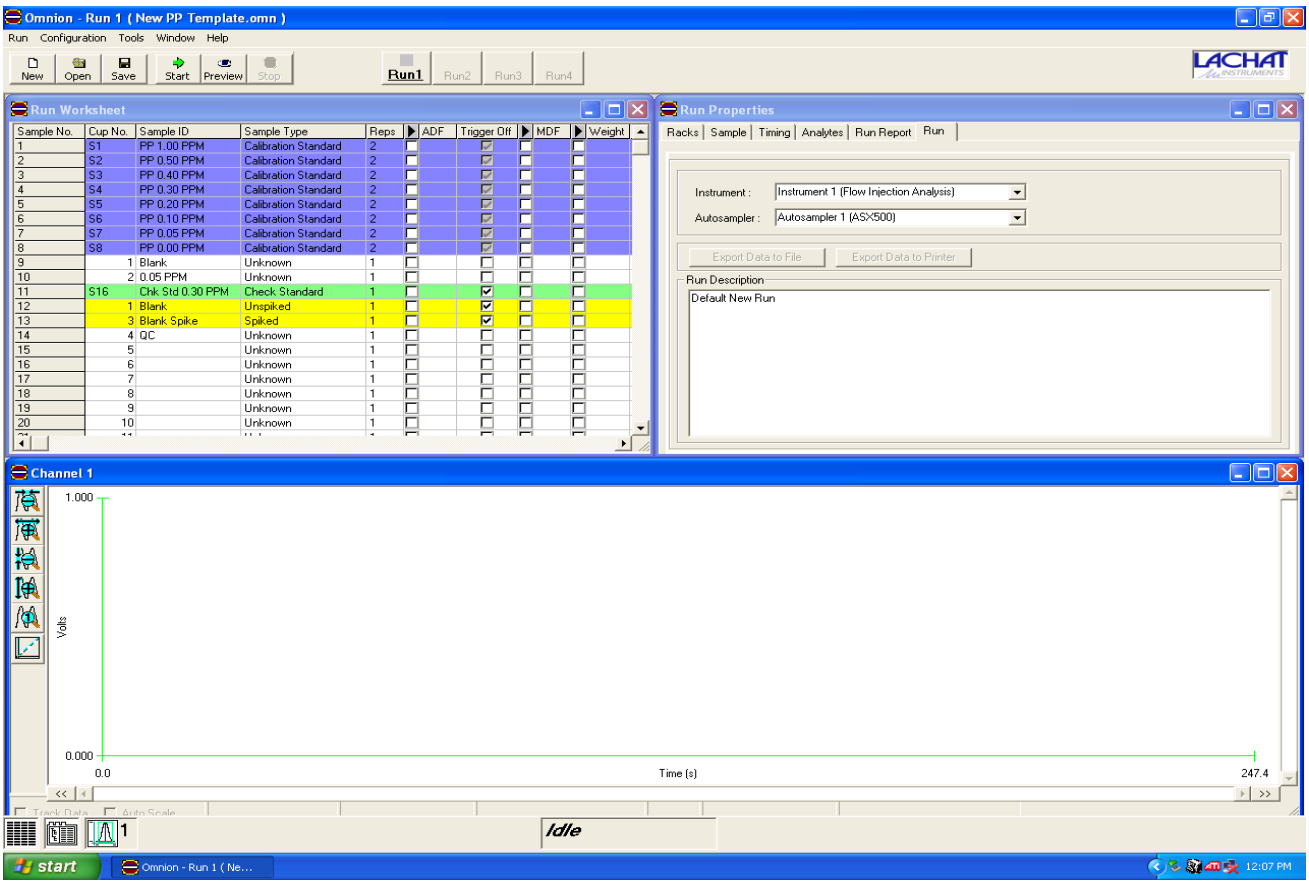
9.1.6 Transfer the filtrate to auto sampler tubes with transfer pipettes.

9.2 Instrument set-up and sample analysis

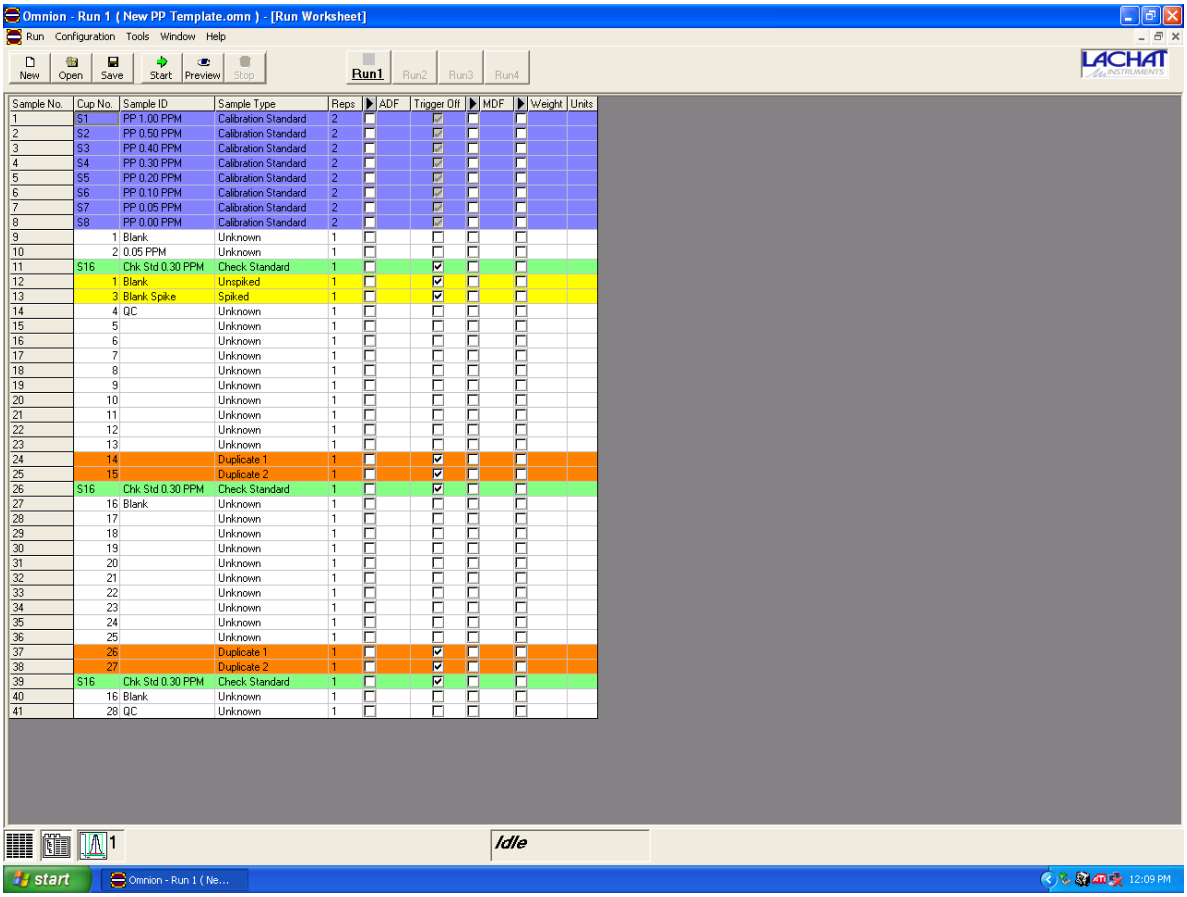
9.2.1 Set up manifold as in the attached diagram.

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.



Sample No.	Cup No.	Sample ID	Sample Type	Reps	ADF	Trigger Off	MDF	Weight	Units
1	S1	PP 1.00 PPM	Calibration Standard	2	☐	☑	☐		
2	S2	PP 0.50 PPM	Calibration Standard	2	☐	☑	☐		
3	S3	PP 0.40 PPM	Calibration Standard	2	☐	☑	☐		
4	S4	PP 0.30 PPM	Calibration Standard	2	☐	☑	☐		
5	S5	PP 0.20 PPM	Calibration Standard	2	☐	☑	☐		
6	S6	PP 0.10 PPM	Calibration Standard	2	☐	☑	☐		
7	S7	PP 0.05 PPM	Calibration Standard	2	☐	☑	☐		
8	S8	PP 0.00 PPM	Calibration Standard	2	☐	☑	☐		
9	1	Blank	Unknown	1	☐	☐	☐		
10	2	0.05 PPM	Unknown	1	☐	☐	☐		
11	S16	Chk Std 0.30 PPM	Check Standard	1	☐	☑	☐		
12	1	Blank	Unspiked	1	☐	☑	☐		
13	3	Blank Spike	Spiked	1	☐	☑	☐		
14	4	QC	Unknown	1	☐	☐	☐		
15	5		Unknown	1	☐	☐	☐		
16	6		Unknown	1	☐	☐	☐		
17	7		Unknown	1	☐	☐	☐		
18	8		Unknown	1	☐	☐	☐		
19	9		Unknown	1	☐	☐	☐		
20	10		Unknown	1	☐	☐	☐		
21	11		Unknown	1	☐	☐	☐		
22	12		Unknown	1	☐	☐	☐		
23	13		Unknown	1	☐	☐	☐		
24	14		Duplicate 1	1	☐	☑	☐		
25	15		Duplicate 2	1	☐	☑	☐		
26	S16	Chk Std 0.30 PPM	Check Standard	1	☐	☑	☐		
27	16	Blank	Unknown	1	☐	☐	☐		
28	17		Unknown	1	☐	☐	☐		
29	18		Unknown	1	☐	☐	☐		
30	19		Unknown	1	☐	☐	☐		
31	20		Unknown	1	☐	☐	☐		
32	21		Unknown	1	☐	☐	☐		
33	22		Unknown	1	☐	☐	☐		
34	23		Unknown	1	☐	☐	☐		
35	24		Unknown	1	☐	☐	☐		
36	25		Unknown	1	☐	☐	☐		
37	26		Duplicate 1	1	☐	☑	☐		
38	27		Duplicate 2	1	☐	☑	☐		
39	S16	Chk Std 0.30 PPM	Check Standard	1	☐	☑	☐		
40	16	Blank	Unknown	1	☐	☐	☐		
41	28	QC	Unknown	1	☐	☐	☐		

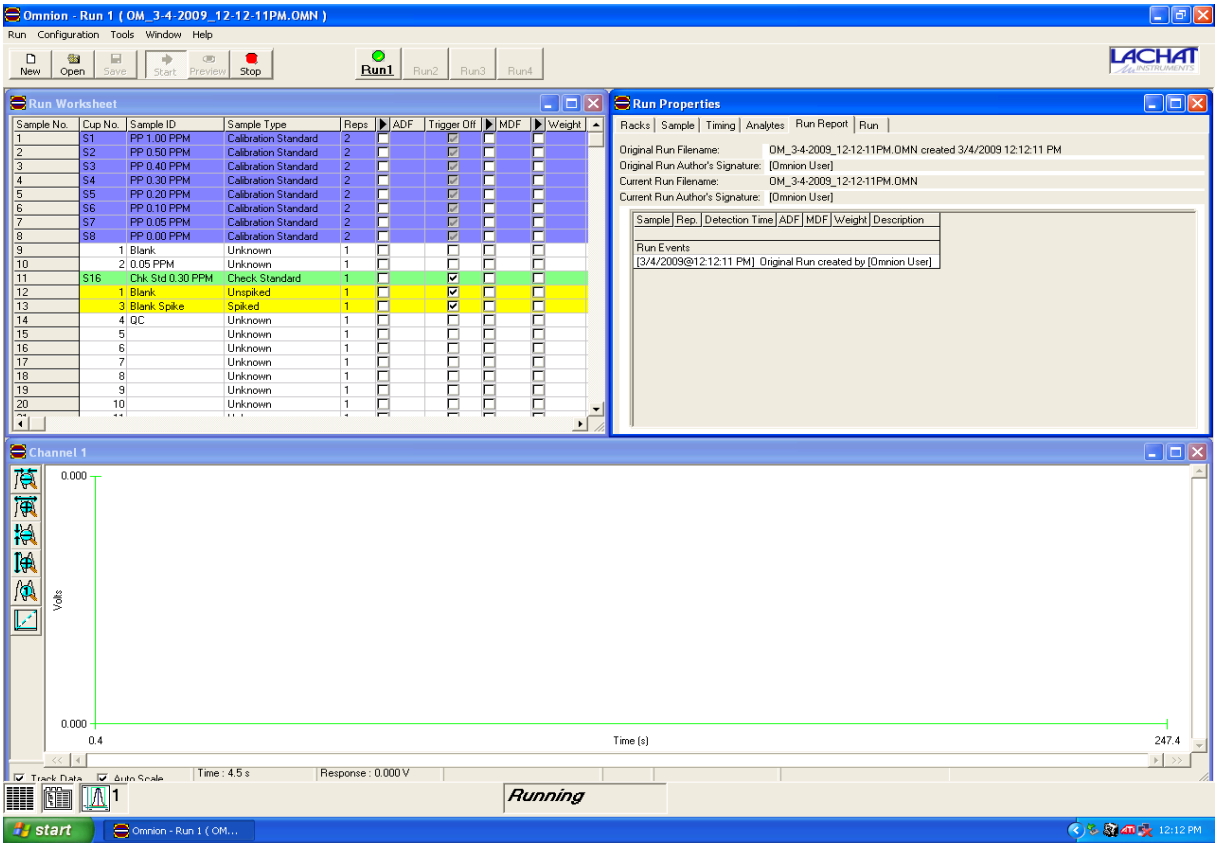
9.2.5 Print a copy of this worksheet by first double clicking on **"Run"** icon and then selecting **"Export Worksheet Data"**.

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!	###
			#DIV/0!	###					#DIV/0!	###
			#DIV/0!	###					#DIV/0!	###
			#DIV/0!	###					#DIV/0!	###
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10.3 Calculate the relative percentage difference for the duplicated samples as follows:

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

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.*

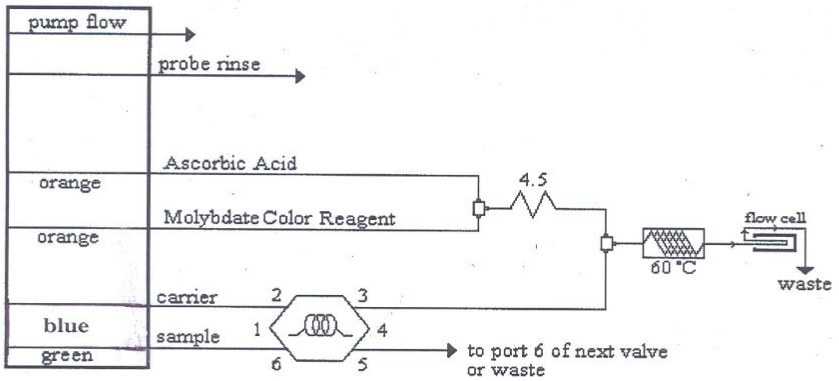
13.4 EPA Method 365.1, *Methods for the Determination of Inorganic Substances in Environmental Samples*, August 1993.

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, Maryland Department of Health and Mental Hygiene, *Quality Assurance Plan,* Revision 11.0, 2011.

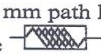
MANIFOLD DIAGRAM FOR PARTICULATE PHOSPHORUS METHOD



Carrier: **Reagent 10**
Manifold Tubing: **0.8 mm (0.032 in) i.d.** This is **5.2 µL/cm.**

QC8000 Sample Loop: 150 cm

Interference Filter: 880 nm

Apparatus: An injection valve, a 10 mm path length flow cell, and a colorimetric detector module is required. The  shows 650 cm of tubing wrapped around the heater block at the specified temperature.

4.5: **70** cm of tubing on a 4.5 cm coil support

APPENDIX A

State of Maryland
DHMH - Laboratories Administration
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		

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

_____ Date Reported: _____

Reviewer's Signature & Date

Reagents ID

1M HCl _____
Ascorbic Acid _____
Color Reagent _____

External QC

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

State of Maryland
DHMH - Laboratories Administration
DIVISION OF ENVIRONMENTAL CHEMISTRY

STANDARD OPERATING PROCEDURES

Title:	Determination of Particulate Carbon and Particulate Nitrogen (Exeter Analytical CE 440)
SOP No.:	IAL-SOP-CE 440/R1.1-11
Revision:	1.1 Replaces: 1.0 Effective: 8/18/2011
Laboratory:	Inorganics Analytical Laboratory
POC:	Moses Obura OburaM@dhmh.state.md.us

Laboratory Supervisor: _____ _____
Signature Date

Division's QA Officer: _____ _____
Signature Date

Division Chief: _____ _____
Signature Date

State of Maryland
DHMH - Laboratories Administration
DIVISION OF ENVIRONMENTAL CHEMISTRY
201 W. Preston Street
Baltimore, MD 21201

REVISION RECORD

Revision	Date	Changes	Made By	Effective Date
0.0	12/09	N/A	Taiyin Wei	1/10
1.0	1/11	New SOP tracking number	Asoka Katumuluwa	
1.1	8/11	Technical and editorial changes	Moses Obura	8/18/11

TABLE OF CONTENTS

STANDARD OPERATING PROCEDURES

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 membranes used in water filtration application.

2.0 SUMMARY OF METHOD

Particulate material on a pre-ignited glass fiber filter is combusted in oxygen helium atmosphere at 900 ^{o}C. The products of combustion are passed over suitable reagents to undergo complete oxidation and removal of undesirable by-products. The mixture then proceeds to a mixing chamber and the uniform gas mixture passes through a series of traps each bracketed with a pair of thermal conductivity detector. 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 ^{o}C and 700 ^{o}C respectively. Wear heat resistant gloves and work on heat resistant bench top when changing these two 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 with Windows XP

5.1.4 Drying oven, 45^{o}C

5.1.5 Muffle furnace, Lindberg

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 Perchlorat - 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 (RPD) is ± 10 %.

8.5 A standard series (blank, 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 ^{o}C for one hour, remove from oven and then place them in a desiccator to be cooled. Remove from the dessicator 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 ^{o}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 ^{o}C until ready for analysis.

9.2.6 Prior to analysis, samples should be placed in a drying oven at 45 ^{o}C for at least 12 hours. Ensure temperature never goes above 50 ^{o}C. Once dried, leave samples in a desiccator until ready to use.

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 conditional, 1 tin capsule blank, 1 conditional, 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 acetalinide.

9.3.2.2 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 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, setting is at 16 psi, oxygen
Pressure at 25 psi, combustion temperature at 900 ^{o}C, and reduction temperature at 700 ^{o}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:

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

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 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, Maryland Department of Health and Mental Hygiene, *Quality Assurance Plan,* Revision 11.0, 2011.

APPENDICES

Appendix A – Data Review Checklist
Appendix B – Sample Run Log
Appendix C – Tube Replacement

APPENDIX A

State of Maryland
DHMH - Laboratories Administration
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 ^{o}C		
Samples Analyzed	Within 5 working days		
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.

_____ Date Reported: _____
Reviewer's Signature & Date

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

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

True Value = _____ Acceptable Range = _____

[3]PN 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 – 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

State of Maryland
DHMH - Laboratories Administration
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

APPENDIX C (continued)

State of Maryland
DHMH - Laboratories Administration
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