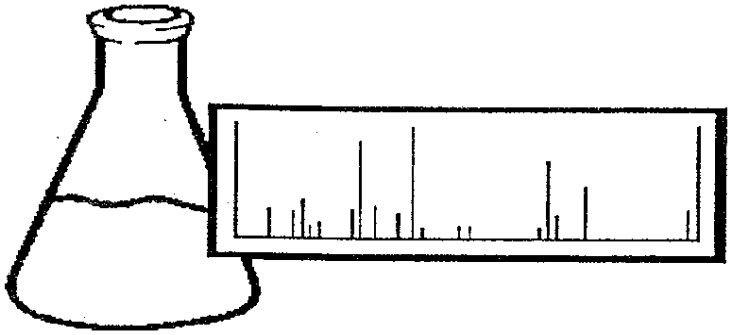
Appendix B

Endyne, Inc. QA/QC Plan
Endyne, Inc. Standard Operating Procedures



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QUALITY SYSTEMS MANUAL

REVISED: March 15, 2001

Revision #: 2

Technical Director: _____ Date : 3/15/01
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Document Control Copy on Buff colored paper. Other Colors not official version

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1.0 Quality Policy Statement

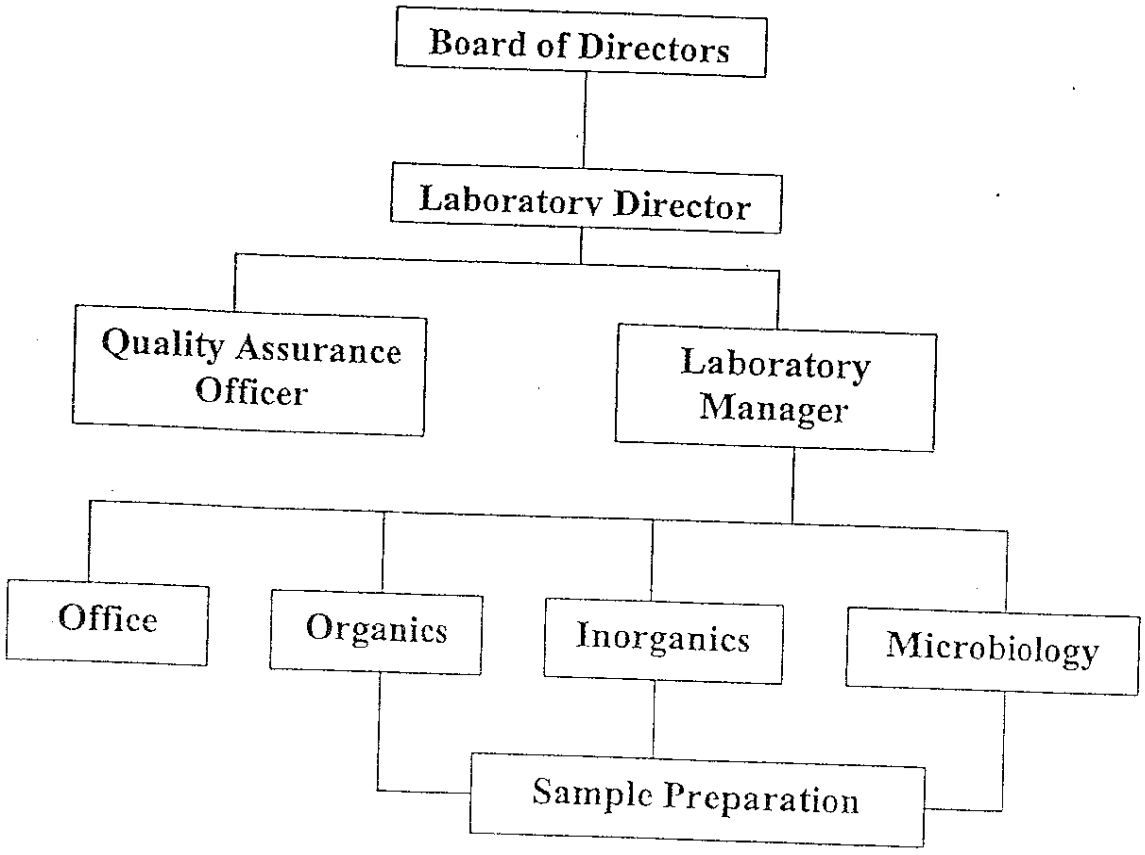
Endyne, Inc. is committed to providing consistent, high quality data in a timely manner. It is our belief that each individual analytical result generated by Endyne is of critical importance to our client. It is thus imperative that Endyne be able to generate and report data of known quality.
To that end, we have outlined our thorough Quality Systems Protocols with the intent of clarifying for our clients all the procedures and precautions that insure the highest level of quality data. Demonstration of Endyne's commitment to maintain our quality objective is evidenced in the following.

- An expertly staffed and fully equipped Laboratory facility.
- Successful participation in multiple EPA/NELAC approved proficiency testing programs.
- Successful implementation of a NELAC compliant quality system.
- Annual internal audits with management review.
- Timely reporting of analytical results.
- Laboratory test results that are supported by quality control data and documented testing procedures.

This quality policy is communicated to all new employees. It is understood, implemented, and maintained by employees at all levels. This is documented by management through the employee evaluation process, the training procedure, the internal audit process, and the document control process.

2.0 Organization Structure/Personnel

The laboratory is comprised of 3 major analytical divisions; Organic, Inorganic and Microbiological. Analysts in each division are trained to perform a variety of analyses and each analysis has at least two analysts trained and qualified to perform the procedure. Laboratory management is comprised of three positions; Laboratory Director, Laboratory Manager and QA Officer. There are two auxiliary divisions which assist in the overall sample flow within the laboratory; Sample Preparation and Office Staff.



2.1 Position Responsibilities

2.1.1 Organic and Inorganic Analysts are responsible for maintaining their own method workloads. Analysts record data and individual QA/QC results in the appropriate databases. Analysts trained in a method perform the initial data quality review of other analysts' work. The QA Officer who reviews the resultant data with the Laboratory Director compiles this information. Analysts report to the Laboratory Manager.

2.1.2 The Chief Microbiologist performs all microbiology and related abilities. The Chief Microbiologist receives training and education necessary to meet certification requirements. The Chief Microbiologist reports directly to the Laboratory Manager.

2.1.3 The Sample preparation department is responsible for all sample preservation and storage at the laboratory. This includes dispersing sample aliquots to the different divisions of the laboratory and ensuring there is sufficient sample volume to perform all requested analyses. Sample technician prepares all sample containers for shipment, including preservation and sampling instructions. The sample tech is also responsible for assigning every sample its unique sample identification number (Reference #).

2.1.4 Office Staff are responsible for the initial receipt of samples at the laboratory, the proper completion of the C-of-C, and the entry of analytical requests into the Laboratory Information Management System (LIMS). Office staff is also responsible for the generation of the final report and ensuring the proper delivery of all analytical results to the client.

2.1.5 The Laboratory Manager coordinates with clients and analysts to develop work schedules to meet analytical demands. The Laboratory Manager is available to analysts to assist in analytical troubleshooting. The Laboratory Manager reports directly to the Laboratory Director.

2.1.6 The Quality Assurance Officer reviews the QA/QC results of individual analyses, compiles the data, and reviews the data with the Laboratory Director. The Q.A. Officer is also responsible for ensuring that the overall laboratory operations comply with the Laboratory QA/QC Plan. The Q.A. Officer reports to the Laboratory Director, but has access to the Board of Directors

2.1.7 The Laboratory Director has overall responsibility for the technical operation of the laboratory. The Director is responsible for arranging and overseeing the maintenance of all equipment and instrumentation, as well as the physical maintenance of the laboratory. The Director provides supervision to all laboratory personnel to ensure adherence to all lab-documented procedures including the Quality Manual.

2.2 Personnel Qualifications

Endyne, Inc. will hire only the most qualified and capable applicants for any position to be filled. Each new employee must have a combination of education and experience to adequately demonstrate the specific knowledge necessary to perform their assigned duties as well as a general knowledge of the mathematics and good laboratory techniques, which would be required.

2.3 Personnel Training

Once hired a new employee will undergo a thorough training regimen to ensure they are capable of performing their tasks to the defined level of acceptance. Proper training consists of an initial introduction to laboratory procedures, continuing general in-house safety and procedural training and where applicable, continuing career development seminars. Most analytical procedures have specific requirements to demonstrate analytical capability. Those requirements are adhered to as well as our general guidelines for training. Documented training activities are stored in employee's personnel files.

2.3.1 Analytical Training

- New analysts review literature associated with the published methods, including internal SOPs and any MSDSs for chemicals used during the procedure. (If it's a new procedure and there is no internal documentation, then the analyst reads the original method, writes a summary and reviews the procedure with the Laboratory Manager.)
- The trainee works several days under the supervision of an analyst, experienced with the method.
- The trainee performs a Demonstration of Capability (DOC). This is an analysis of four replicate quality control samples. The average of the results must be within 20% of the target value and the standard deviation must be less than 20%, unless the method specifically indicates otherwise.
- The Laboratory Manager will authorize an analyst to perform a method only after successful completion of the DOC and upon conference with the primary trainer. Documentation of this authorization will be kept in the employee's personnel file in the form of an "Analyst Certification Form".

2.3.2 General Procedural and Ethical Training

2.3.2.1 Initial Training

- An overall review of sample flow through the laboratory is provided, so that they may better understand their relationship within the laboratory.
- A review of safety protocols and emergency response procedures is performed.
- A review of general laboratory procedures, including balance, pipetting, etc. techniques is performed.
- Ethical and legal responsibilities are outlined, including potential punishments and penalties for improper, unethical or illegal actions.

2.3.2.2 Continued Training

- Weekly Inorganic and Organic department meetings are held to discuss safety and quality control issues that arise during the course of the previous week.
- Laboratory wide staff meetings occur as needs dictate. Topics range from employee benefits and staffing changes to analytical issues and ethical/legal responsibilities.
- Employees attend off-site training seminars, which are deemed valuable by management. These have included office skills, hazardous waste handling, safety training, and analytical instrumentation courses.

3.0 Laboratory Environment

Endyne, Inc. consists of three levels covering approximately 2500 square feet of work area. The top floor consists of meeting rooms and organic prep/extraction lab. The middle floor consists of office space and the Organics instrumentation lab. The bottom floor consists of the Microbiology Lab and the Metals and Inorganics work areas.

Endyne has no equipment requiring extremely tight environmental conditions. Each floor contains its own heating and cooling systems designed to keep temperatures around 70°C and humidity around 40%. Electronic scales are mounted on vibration inhibiting platforms. Exhaust hoods are located on each floor. They are inspected and calibrated on an annual basis

Biological work areas are sterilized between uses. Test areas of incompatible activities are separated to eliminate contamination. Work areas are wiped down at the end of each day and good housekeeping practices in general are followed to minimize contamination and health risks.

4.0 Document Control.

The Document Control System is designed so that documents, which are generated by the laboratory and deemed critical to proper laboratory operation, are tracked and access is limited to responsible parties. This system ensures that only the most recent revisions are available to the appropriate personnel, and all documents in use have the proper authorization. Documents covered by this system are listed in the Master Document Control List, which lists all the records, their location, and revision history. Records covered in the Document Control List include the following;

-The Master Document Control List
-This Quality Assurance Manual
-All Analytical SOPs
-Equipment Maintenance SOPs, i.e. Analytical Balance and Pipette SOPs
-General Laboratory Procedure SOPs, i.e. Mailing and Billing procedures
-Sampling Procedures, i.e. Coliform Bacteria
-Employee Qualification Summaries
-AIHA Laboratory related documents

The Quality Assurance Officer is responsible for maintaining the Document Control System. Master List Controlled Documents are indicated by the paper color indicated in the footer. Uncontrolled copies are determined by being printed on any other colored paper. The Q.A. Officer has control over the supply of the specified colored paper. The Laboratory Director approves all newly released control documents. The Q.A. Officer is responsible for archiving the obsolete documents.

5.0 Sample Management

5.1 Review of all New Work

All new work is defined as analytical procedures for which Endyne does not already have an internal documented procedure with established quality control objectives. A list of established analyses is provided in Table 2. All new work is accepted by the Laboratory Director or Laboratory Manager, after ensuring the laboratory has the proper facilities and equipment to perform the analyses. Staff, involved in the new work, meets to discuss any pertinent issues regarding quality assurance issues. The plan for any new testing must be reviewed and approved by the Laboratory Director. The designated employee(s) shall write a SOP for the procedure, which will be included in the document control. A DOC will be kept in the employees personnel file.

5.2 Sampling Protocol

In general, samples received by Endyne are collected by representatives of firms contracting with the laboratory to perform the analysis. These firms are responsible for insuring that proper sampling procedures are followed. Endyne's responsibility is limited to providing written sampling procedures for sample collection, preservation, field filtration and transport for specific analysis when requested. Upon request, Endyne can provide expert sampling services for air, water, and solid media.

5.3 Sample Receipt Protocol

Upon arrival at the laboratory, samples and associated Chain-of-Custody (C-of-C) are reviewed for accuracy and sample integrity. In summary, the C-of-C should contain the following information:

1. Client and contact person.
2. Project Name
3. Client sample identification (station location or number).
4. Name of sample collector, date and time of collection.
5. Analyses requested.
6. Container type and volume.
7. Type of preservation performed in the field.
8. Special requests or instructions (e.g. rush analyses).
9. Name of Endyne personnel who received samples.
10. Date and time of receipt.

Samples are then inspected for general condition, which includes the following:

1. If seals are present on containers, an inspection of each seal is made to insure that each seal is intact.
2. Proper sampling containers have been utilized for the requested analyses, and adequate sample volume is available.
3. For volatile organic compounds (VOC), any air bubbles will be noted on the C.O.C.
4. Proper temperature and preservation guidelines have been followed.
5. Comparison of notations on sample containers with those on the Chain-of-Custody (sample location, date, time) are made to insure that all samples which were collected have arrived at the laboratory, and are properly labeled.

If discrepancies or omissions exist in any of these areas, the Laboratory Director and client are notified. If the client cannot be reached, the samples are placed in cold storage (4°C) and no further action is taken until the problem is resolved.

5.3 Sample Receipt Protocol (cont.)

In conjunction with the C-of-C, Endyne uses a Sample Prep Report Form to document custodial and sample condition issues. The person receiving the sample records the condition of the samples, the temperature, the accuracy of the C-of-C and the means of delivery to the laboratory. The person prepping the sample records any preservation or sample preparation procedures necessary for sample storage until the time of analysis.

Once logged in, the samples are delivered to the appropriate department, preserved, as appropriate, and refrigerated at 4°C.

5.4 Chain-of-Custody

The purpose of the Chain-of-Custody form is to provide a written record, which traces the collection, possession, and handling of samples from the moment of collection to arrival at the laboratory. An example of our Chain-of-Custody Record is provided in Figure 1. The Chain-of-Custody form is signed by the sample collector, by those responsible for the transportation of the sample, and finally, by laboratory personnel. Any breach in the Chain-of-Custody is reported to the Laboratory Director, the sampler, and the client. In most instances, the Chain-of-Custody form will also be used by the client as an order form to specify the requested analyses, and as a reporting sheet for field results. Upon receipt at the laboratory, a carbon copy of the Chain-of-Custody form is given to the sampler. All original forms become property of the laboratory.

5.5 Sample Preservation

Sample preservation is divided into two areas. They are the following:

1. Field preservation by sampler.
2. Preservation by Endyne personnel.

Endyne, Inc. takes no responsibility for preservation procedures or materials used in the field. As stated in Section 5.2, we will provide methods, bottles and chemicals appropriate for proper field preservation. Any observed deviation of field preservation from these recommended methods will be reported to the client and noted on the Chain of Custody.

If the sample is received without preservation, Endyne personnel will preserve the sample following Table 1, as long as handling the sample will not affect data integrity. Any non-standard preservation requested by the sampler will be reported to the client and noted on the Chain-of-Custody.

5.6 Analytical Holding Times

Also listed on Table 1 are recommended holding times for all analyses. As discussed in Section 5.3, notification will be made to the Laboratory Director of any samples received by Endyne, which have exceeded the specified holding time limit. At Endyne, all samples will be processed within the recommended holding time. If, due to equipment malfunction or other circumstances beyond the control of the laboratory, an analysis cannot be performed at Endyne, a subcontracting laboratory will be assigned to perform the analysis. A Chain-of-Custody record will be forwarded with all samples sent to the subcontracting laboratory.

5.7 Minimum Required Volumes

Table 1 indicates the minimum required volumes to perform all analyses. During laboratory preservation procedures (see Section 5.5), the sample will be split, if necessary, such that appropriate volumes are preserved for all requested analyses. The notification procedure set forth in Section 5.3 will be implemented for samples with inadequate volume. In these cases, it will be the client's responsibility to decide which analyses to perform.

6.0 Instrumentation and Equipment

The proper operation of the analytical instrumentation, all measuring devices, the de-ionized water supply, as well as the cleanliness of the glassware must be insured to meet the quality control requirements of the laboratory. Individual Standard Operating Procedures are maintained and adhered to for each type of Equipment. The following sections are summaries of those SOPs. A listing of all major pieces of equipment is provided in Table 3.

6.1 De-Ionized Water

A continuous resistivity meter is mounted on the de-ionized water unit. De-ionized water has an acceptable quality when the resistance value ranges from 14 to 18 μohms. If the resistance falls below this value, new filter elements are installed. De-ionized water from the conditioned device is not used until it can pass the 14 to 18 μohms standard. Grab samples are also analyzed using a separate electronic conductivity meter on a monthly basis, to verify the accuracy of the continuous recording device.

6.2 Balances, Thermometers and Pipettors

All weights and thermometers are calibrated against NIST traceable standards at an offsite certified facility. Balances are inspected by a certified specialist on an annual basis and an internal calibration is performed at a minimum of once a week. A variety of pipettors capable of measuring from 10ul to 10ml are calibrated on a monthly basis

6.3 Glassware

All glassware used for measurement is Class A certified. Glassware is cleaned thoroughly with soap and water. Tap water is used for the preliminary wash followed by rinses with de-ionized water. For trace metal analysis, the glassware is also subjected to an acid wash. All glassware used for organic extraction is rinsed with appropriate solvent prior to analysis.

6.4 Instrumentation

Each major piece of instrumentation is assigned to an individual analyst, for which they are responsible. A separate bound notebook is maintained at each instrument station. In this notebook, the daily operation status of the instrument is noted as well as its complete maintenance history. All changes and servicing to the original factory equipment are noted, dated, and signed by the analyst responsible for the instrument. The Laboratory Director is notified of any problems with the equipment. Routine checks of the instrument log notebooks are made by the Laboratory Director to insure proper reporting.

Regular site visits are made to Endyne by manufacturer's service staff for equipment servicing. Some major instruments are covered by service contracts, which provide for rapid response by the manufacturer in the event of an equipment malfunction.

7.0 Internal Quality Control of Test Procedures

All method-referenced analyses performed at Endyne adhere to the quality control standards outlined in the published literature. Methods, which do not specifically reference quality control parameters, as well as in-house developed methods, adhere to, at the minimum, the QA/QC protocols outlined here. In some cases however, some of the following protocols are not possible to perform. Individual method SOPs have the appropriate QA/QC protocols referenced within them.
Endyne's internal QA/QC plan consists of method and analyst capability demonstrations, as well as a suite of quality control protocol steps during the analysis of samples.

7.1 Method and Analyst Capability Demonstrations

All records include references to the raw data files or dates analyzed, so that calculations can be regenerated from the original data.
Demonstration of Capability (DOC)- Every new analyst performs a set of four mid-range concentration LFBs. The LFBs should be prepared as one sample, then split into four aliquots. The percent standard deviation and average percent recovery are determined for each analyte. Each must be less than 20% to ensure proper capability of the analyst for that method. Documentation of the DOC is kept in the analyst's personnel file.
Method Detection Limit Study (MDL)- The analyst performs a set of seven LFBs at a concentration of 2 to 5 times the expected MDL value. The standard deviation is multiplied by 3.14 to determine the laboratories MDL. The MDL should be less than half the laboratory's reporting limit. MDLs are performed on an annual basis at the beginning of each year. Copies of the MDL are stored with the QA/QC data for the method as well as with the method SOP for that year.

7.2 Quality Control Protocols

Endyne generates control limits for the quality control samples using method-defined limits. Typically, those limits are +/- three standard deviations of the average recovery. In the event there is no accrued history of QC data an absolute acceptance limit of 80%-120% is employed. Duplicate precision limits are based on 3.27 multiplied by the mean relative percent differences. In the event, there is no accrued history of duplicate data an absolute acceptance limit of 20% relative percent difference.

Analytical Calibration Line- A minimum of a 3-point calibration line not including "0", which must have a linear correlation of 0.995 or better. The lowest point in the line must be less than the reporting limit or regulatory level.
Continuing Calibration Check (CCC)- A calibration line standard is analyzed if a calibration line already exists. A CCC is analyzed at the start of each analytical day.
Laboratory Reagent Blank (LRB)- A reagent water sample which is treated as a normal sample and must be shown to be free of all target analytes at reportable levels. An LRB is run with every 20 samples or each analytical batch, whichever is more frequent.
Laboratory Fortified Blank (LFB)- An independent source standard containing all the target compounds is analyzed like a normal sample. A LFB is run with every 20 samples or each analytical batch, whichever is more frequent.
Matrix Duplicate (MD)- Aliquots of the same sample which are analyzed identically. A MD is run with every 20 samples or each analytical batch, whichever is more frequent
Matrix Spike (MS)- A sample aliquot to which a known amount of analyte(s) is added. A MS is run with every 20 samples or each analytical batch, whichever is more frequent.
Matrix Spike/Duplicate (MS/D)- This quality control parameter may be employed in place of the Sample Duplicate and Matrix Spike. Two sample aliquots are spiked with known amounts of analyte and the percent recovery and percent difference is determined for the samples.

7.3 Analytical Standards

Endyne takes special care to ensure the accuracy of all analytical standards. Standards are made in-house from neat materials or purchased from suppliers with at least an ISO 9001 certification. All Certificates of Analysis from suppliers are kept on file in the department in which they are used. In-house standards are made using Class A glassware and a NIST traceable balance. All working standards are documented in individual method logbooks that insure traceability to the original manufacturer.

7.4 Data Storage and Archival

All raw data and calculations, sufficient to completely reconstruct the data packet, are kept on file by the laboratory for a period of 5 years. Data storage during the course of the year is outlined in individual SOPs. At the start of each calendar year all data and paperwork is archived in banker boxes. These boxes for the two previous years are kept on site. Data older than two years is kept off-site in a secured location.

7.5 Internal Audit and Data Review

-Raw Data - Each analyst must keep detailed information of the raw data in a bound notebook or on the computer printout. Entries must include the analyst initials, date and time of analysis, sample identification number, parameter tested and analytical method, documentation of all readings, and results of all weightings and dilutions. All calculations documenting the final concentration of the constituent as well as determinations of all quality control samples must be recorded with the data.

-Data Checking – Data checking is initially performed by an independant analyst fully trained in the analytical methodology. After the analyst has processed all the associated data and recorded the results of an analysis, the independent analyst reviews the raw data and associated quality control samples. The reviewer then signs off on the Log-In report form indicating that they have fully reviewed the work. The completed report is submitted to the Laboratory Director for final review and approval before being released to the client.

-System Audits – The QA Officer and/or the Laboratory Director will review individual methods on an annual basis with the primary analyst. Procedures will be compared to the SOP and Quality Systems Manual to confirm adherence to both. Any corrective actions will be documented. Where audit findings cast doubt on the validity of results, clients will be contacted immediately in writing.

-Managerial Review – On an annual basis, the Laboratory Director will review the quality system and its testing and calibration activities. The review will be based on feedback from the QA Officer, the results of proficiency tests, as well as audits from external certification officers. Any necessary changes will be implemented at this time and documented in the Quality Systems Manual.

8.0 Testing Discrepancies and Corrective Action

Occasionally analytical data fails the quality control protocols outlined in each method SOP. Procedures for determining the source of these failures are outlined in the SOP. In general, the primary analyst immediately reviews the work and assesses where the error occurred, corrects the deficiency and reanalyzes the associated samples. No further sample analysis is performed until the source of the error is determined and corrected. The laboratory manager and director are available for immediate consultation if necessary. In some instances, due to the sample volume submitted or the expiration of the analytical holding time, it is impossible to reanalyze the sample within QA/QC protocols. In that event the client is notified of the violation by telephone or in writing. No data, which is outside method specified quality control parameters, will be reported without the appropriate notation informing the client of the transgression.

9.0 Exceptionally Permitted Departures from Documented Policies

The Laboratory Director is responsible for ensuring adherence to the documented policies of the laboratory. However, in some instances departure from documented procedure may be necessary. In this instance, digression from the procedure is allowed as long as the changes are fully documented as well as the reason why the deviation occurred and the Laboratory Director approves all the departures.

10.0 Proficiency Testing

Endyne participates in several proficiency-testing programs for drinking water, wastewater and air. Endyne participates in a Water Supply testing program on a semi-annual basis. The testing encompasses all of the parameters for which Endyne receives certification for from the State of Vermont. Endyne also participates in semi-annually waste water prficiency testing administered by the New York State Environmental Laboratory Accreditation Program (ELAP). Both sets of proficiency testing are approved by the National Environmental Laboratory Accreditation Council (NELAC). Endyne also participates quarterly in a suite of testing for in-door air contaminants. These tests are administered by the American Industrial Hygienists Association (AIHA).

11.0 Complaints and Corrective Action Procedures

Endyne continually strives to improve data quality and customer satisfaction. In an effort to track complaints and quality control problems, Endyne maintains a file where complaints, from clients or other parties, and their corrective actions are recorded. The QA Officer immediately investigates the affected areas of the laboratory and submits a written report, describing the problem and the appropriate corrective action, to the Laboratory Director. The issue is then discussed during the weekly department meetings with everyone who may encounter a similar problem. If the issue is a data quality matter, no additional data is reported until the matter is resolved.

12.0 Reporting of Results

Endyne policy is to report results solely to the client listed on the Chain-of-Custody. Delivery of results is accomplished in three ways. The only authoritative results reported by Endyne are in the form of a hard copy report with the original signature of the Laboratory Director or appointed signer. The other forms of data deliverables are by fax or e–mail. Clients who request a fax must indicate on the C-of-C whom will receive results or if the request is made by phone proof must be made that they are the stated client. This is usually accomplished by referencing the distinctive C-of-C sequence number. Deliverables in the form of e-mails are only provided to established customers, whom have made formal arrangements for receipt of results.

13.0 Revision History

July 18, 2000:	Complete Draft
March 15, 2001:	Minor Revision

14.0 References

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CHAIN-OF-CUSTODY-RECORD

37472

Project Name:		Reporting Address:	Billing Address:
Endyne Order ID: (Lab Use Only)	-O -I -S	Company: Contact Name/Phone #:	Sampler Name: Phone #:

Ref # (Lab Use Only)	Sample Identification	Matrix	GRAB	COMP	Date/Time	Sample Containers No.	Sample Containers Type/Size	Field Results/Remarks	Analysis Required	Sample Preservation	Rush

Relinquished by:	Date/Time	Received by:	Date/Time	Received by:	Date/Time

New York State Project: Yes ____ No ____

Requested Analyses

1	pH	6	TKN	11	Total Solids	16	Sulfate	21	1664 TPH/FOG	26	8270 PAH
2	Chloride	7	Total P	12	TSS	17	Coliform (Specify)	22	8015 GRO	27	PP13 Metals
3	Ammonia N	8	Total Diss. P	13	TDS	18	COD	23	8015 DRO	28	RCRA8 Metals
4	Nitrite N	9	BOD	14	Turbidity	19	8021B	24	8260/8260B	29	
5	Nitrate N	10	Alkalinity	15	Conductivity	20	8010/8020	25	8270 B/N or Acid	30	
31	Metals (As Is, Total, Diss.) Ag, Al, As, B, Ba, Be, Ca, Cd, Co, Cr, Cu, Fe, Hg, K, Mg, Mn, Mo, Na, Ni, Pb, Sb, Se, Si, Sr, Ti, Tl, V, Zn										
32	TCLP (Specify: volatiles, semi-volatiles, metals, pesticides, herbicides)					33					
34	Other										

(White, Yellow, Pink Copy - Laboratory / Goldenrod Copy - Client)

TABLE 1
Recommendation for Sampling and Preservation of Samples According to Measurement

Measurement	EPA Method	Vol. Req. (ml)	Container[2]	Preservative[3,4]	Holding Time[5]
100 PHYSICAL PROPERTIES					
Color	110.3	100	P,G	Cool, 4°C	48 hrs.
Conductance	120.1	100	P,G	Cool, 4°C	28 days
Hardness	6010	100	P,G	HNO_3 to pH <2	6 mos.
Odor	140.1	500	G only	Cool, 4°C	24 hrs.
pH	150.1	25	P,G	None Required	Analyze Immediately
Residue/Solids:					
Filterable	160.1	250	P,G	Cool, 4°C	7 days
Non-filterable	160.2	250	P,G	Cool, 4°C	7 days
Total	160.3	250	P,G	Cool, 4°C	7 days
Volatile	160.4	250	P,G	Cool, 4°C	7 days
Settleable Matter	160.5	1000	P,G	Cool, 4°C	48 hrs.
Temperature	170.1	100	P,G	None Required	Analyze Immediately
Turbidity	180.1	100	P,G	Cool, 4°C	48 hrs.
200 METALS					
Dissolved	3113B/200.7/6010	500	P,G	Filter on-site HNO_3 to pH <2	6 mos.
Suspended	3113B/200.7/6010	500	P,G	Filter on-site	6 mos.
Total	3113B/200.7/6010	500	P,G	HNO_3 to pH <2	6 mos.
Chromium^{+6}	7196A	200	P,G	Cool, 4°C	24 hrs.

TABLE 1
Recommendation for Sampling and Preservation of Samples According to Measurement

Measurement	EPA Method	Vol. Req. (ml)	Container[2]	Preservative[3,4]	Holding Time[5]
300 INORGANICS, NON-METALLICS					
Acidity	305.1	100	P,G	Cool, 4°C	14 days
Alkalinity	310.1	100	P,G	Cool, 4°C	14 days
Bromide	300.0	100	P,G	None Required	28 days
Chloride	300.0	50	P,G	None Required	28 days
Chlorine	330.5	200	P,G	None Required	Analyze Immediately
.Cyanides	335.1	250	G	Cool, 4°C NaOH of pH >12 0.6 g ascorbic acid	14 days
Fluoride	340.2	200	P,G	None Required	28 days
Nitrogen:					
Ammonia	350.3	100	P,G	Cool, 4°C $NaSO_4$ to pH <2	28 days
Kjeldahl, Total	351.4	100	P,G	Cool, 4°C $NaSO_4$ to pH <2	28 days
Nitrate	300.0	50	P,G	Cool, 4°C	48 hours
Nitrite	300.0	50	P,G	Cool, 4°C	48 hours
Phosphorus:					
Ortho-Phosphate,	365.2	100	G	Cool, 4°C H_2SO_4 to pH <2	28 days.
Dissolved	365.2	100	G	Filter,Cool, 4°C H2SO4 to pH <2	28 days
Total	365.2	150	G	Cool, 4°C H_2SO_4 to pH <2	28 days
Total Dissolved	365.2	50	G	Filter on-site Cool, 4°C H_2SO_4 to pH <2	24 hrs.

TABLE 1
Recommendation for Sampling and Preservation of Samples According to Measurement

Measurement	EPA Method	Vol. Req. (ml)	Container[2]	Preservative[3,4]	Holding Time[5]
300 INORGANICS, NON-METALLICS					
Sulfate	300.0	50	P,G	Cool, 4°C	28 days
Sulfide	376.1	500	P,G	Cool, 4°C Add 2ml Zinc Acetate and NaOH to pH >9	28 days
Sulfite	377.1	50	P,G	None Required	Analyze Immediately
Radioactivity:					
Gross Alpha	Rad TSU	2000	P	None	28days
Radium 226,228	Rad TSV	4000	P	None	28 days
400 ORGANICS					
BOD	405.1	1000	P,G	Cool, 4°C	48 hrs.
BOD, Soluble	405.1	1000	P,G	Cool, 4°C	48 hrs
COD	410.1	50	P,G	Cool, 4°C H_2SO_4 to pH <2	28 days
Organic Carbon	415.1	25	P,G	Cool, 4°C H_2SO_4 or HCl to pH <2	28 days
Phenolics	420.1	500	G only	Cool, 4°C H_2SO_4 to pH <2	28 days
MBAs	425.1	1000	P,G	Cool, 4°C	48 hrs.
Oil & Grease	1664	1000	G only	Cool, 4°C H_2SO_4 to pH <2	28 days

TABLE 1
Recommendation for Sampling and Preservation of Samples According to Measurement

Measurement	EPA Method	Vol. Req. (ml)	Container[2]	Preservative[3,4]	Holding Time[5]
500 SERIES Drinking Water					
Volatiles	504.1	2 40-ml vials	G only	Cool, 4°C 10 mg $Na_2S_2O_3$	14 days
Pesticides/PCB	505	2 40-ml vials	G only	Cool, 4°C 10 mg $Na_2S_2O_3$	14 days/
Herbicides	515	2 1000-ml	Amber G	4°C Preservation in Lab	14 days/ 14 days
S.O.C.	525	2 1000-ml	Amber G	4°C Preservation in Lab	14 days/
Carbamates	531	2 40ml vials	G only	Cool, 4°C 1.8mls MCA	28 days
V.O.C	524.2 503/502	2 40ml vials	G only	Cool, 4°C 1:1 HCl	14 days
8000 SOLIDS					
Volatiles	8010/8021B 8260/8260B	Full 40 ml vial	G only	4°C	14 days
Pesticides	8081A	250 ml	Amber G	4°C	14 days/ 40 days
PCBs	8082	250 ml	Amber G	4°C	14 days/ 40 days
S.V.O.C. BN/A	8270C	250 ml	Amber G	4°C	14 days/ 40 days
TPH GRO	8015B	2 40-ml	G only	4°C to pH <2	14 days
TPH DRO	8015B	2 40-ml	G only	4°C to pH <2	14 days 40 days

TABLE 1
Recommendation for Sampling and Preservation of Samples According to Measurement

Measurement	EPA Method	Vol. Req. (ml)	Container[2]	Preservative[3,4]	Holding Time[5]
8000 WATER					
Volatiles	8010/8020 8260/8260B	2 40-ml vials	G only	No bubbles 4°C HCl to pH <2	14 days
Pesticides	8081A	2 1000-ml	Amber G	4°C pH between 5-9	7 days/ 40 days
PCBs	8082	2 1000ml	Amber G	4°C pH between 5-9	7 days/ 40 days
S.V.O.C. BN/A	8270C	2 1000-ml	Amber G	4°C	7 days/ 40 days
TPH GRO	8015B	2 40-ml	G only	No bubbles 4°C HCl to pH <2	14 days
TPH DRO	8015B	2 40-ml	G only	No bubbles 4°C HCl to pH <2	14 days
MICROBIOLOGICAL					
Total and Fecal Coliform	Not Applicable	100 ml	125 ml Sterile Plastic	4°C 0.008% Na2s203	30 hrs (24 hrs, recommended)
Escherichia coli	Not Applicable	100 ml	125 ml Sterile Plastic	4°C	24 hrs
Heterotrophic	Not Applicable	100 ml	125 ml Sterile Plastic	4°C	8 hrs

TABLE 1 FOOTNOTES

1. Referenced Methods are from approved publications from Standard Methods 17th Edition, Solid Waste 846 and Environmental Protection Agency

2. A general discussion on sampling water and industrial wastewater may be found in ASTM, Part 31, pgs. 72-82 (1976) Method D-3370.

3. Plastic (P) or Glass (G). For metals, polyethylene with a polypropylene cap (teflon liner is preferred.

4. Sample preservation should be performed immediately upon sample collection. For composite samples each aliquot should be preserved at the time of collection. When use of an automated sampler makes it impossible to preserve each aliquot, then samples may be preserved by maintaining at 4°C until compositing and sample splitting is completed.

5. When any sample is to be shipped by common carrier or sent through the United States Mail, it must comply with the Department of Transportation Hazardous Materials Regulations (49 CFS Part 172). The person offering such material for transportation is responsible for ensuring such compliance. For the preservation requirements of Table 1, the Office of Hazardous Materials, Materials Transportation Bureau, Department of Transportation has determined that the Hazardous Materials Regulations do not apply to the following materials: Hydrochloric acid (HCl) in water solutions at concentrations of 0.04% by weight or less (pH about 1.96 or greater); Nitric acid (HNO_3) in water solutions at concentrations of 0.15% by weight or less (pH about 1.15 or greater); Sulfuric acid (H_2SO_4) in water solutions at concentrations of 0.35% by weight or less (pH about 1.15 or greater); Sodium hydroxide (NaOH) in water solutions at concentrations or 0.080% by weight or less (pH about 12.3 or less).

6. Samples should be analyzed as soon as possible after collection. The times listed are the maximum times that samples maybe held before analysis and still considered valid. Samples may be held for longer periods only if the permittee , or monitoring laboratories, has data on file to show that the specific types of sample under study are stable for the longer time, and has received a variance from the Regional Administrator. Some samples may not be stable for the maximum time period given in the table. A permittee, or monitoring laboratory, is obligated to hold the sample for a shorter time if knowledge exists to show this is necessary to maintain sample stability.

7. Should only be used in the presence of residual chlorine. The sample is filtered and the NaOH is added to pH 12.

8. For samples from non-chlorinated drinking water supplies, concentrated H_2SO_4 should be added to lower sample pH to less than 2. The sample should be analyzed before 14 days.

TABLE 2 Analytical Methods
Organic Analyses

ANALYTE	EPA METHODS
Volatile Organic Compounds	
VOCs by GC/MS	8260/8260B
GC/MS Calibrated Nontarget Compounds	8260B
SDWA Drinking Water Volatiles	524.1
Purgeable Aromatics by GC/PID	8020/8021B
Purgeable Halocarbons by GC/MS	8010
Combined Purgeables by GC/MS	8010&8020
SDWA EDP, DBCP by GC/ECD	504.1
Base Neutral/Acid Extractables	8270C
Acid Extractables	8270C
Base Neutral Extractables	8270C
Polynuclear Aromatics	8270C
Organochlorine Pesticides & PCBs	8081A
PCBs	8082
Herbicides	8150
SDWA Pesticides	505/525.2
SDWA PCBs	505
SDWA Herbicides	515.2
SDWA Carbamates	531.1
Anti-Freeze	8015B
TPH-GRO	8015B
TPH-DRO	8015B/1664
Petroleum Hydrocarbon Identification	GC/FID
Oil&Grease	1664
Oil and Grease by IR	413.2
Petroleum Hydrocarbons by IR	418.1

TABLE 2 Analytical Methods
Inorganic Analyses

PARAMETER	EPA METHODS
Alkalinity	310.1
Biological Oxygen Demand	405.1
Carbon:	
Total Inorganic (water)	415.1
Total Organic (water)	415.1
Total (soil)	415.1
Total Organic (soil)	415.1
Chemical Oxygen Demand	410.2
Chloride	325.1
Chlorine	330.5
Color	110.3
Cyanide - Total	335.2
Cyanide - Total & Amenable	335.1
Fluoride	340.2
Hardness	SM314A
Halides, Total Organic (TOX):	
Water	9020
Solids	9020
Lime Requirement	----
Nitrogen	
Ammonia	350.3
Nitrate	300.0
Nitrite	300.0
Total Kjedahl	351.4

TABLE 2 **Analytical Methods**
Inorganic Analyses

PARAMETER	EPA METHODS
pH	150.1
Phenols - Total	420.3
Phosphorus:	
Total	365.2
Ortho	365.2
Total Dissolved	365.2
Solids	
Total	160.3
Percent	160.3
Suspended	160.2
Dissolved	160.1
Volatile	160.4
Suspended Volatile	160.2/160.4
Settleable	160.5
Specific Conductance	120.1
Sulfate	300.0
Sulfide	376.1
Sulfite	377.1
Surfactants - MBAs	425.1
Turbidity	180.1

TABLE 2 **Analytical Methods**
Metal Analyses

ANALYTE	EPA METHODS
Aluminum	200.7/6010
Antimony	200.7/6010
Arsenic	200.7/3113B
Barium	200.7/6010
Beryllium	200.7/6010
Boron	200.7/6010
Cadmium	200.7/3113B
Calcium	200.7/6010
Chromium	200.7/6010
Colbalt	200.7/6010
Copper	200.7/6010
Iron	200.7/6010
Lead	200.7/3113B
Magnesium	200.7/6010
Manganese	200.7/6010
Mercury (cold vapor)	245.1/7470
Molybdenum	200.7/6010
Nickel	200.7/6010
Potassium	200.7/6010
Selenium	200.7/3113B
Silver	200.7/6010
Sodium	200.7/6010

TABLE 2 Analytical Methods Metal Analyses	
Thallium	200.7/3113B
Vanadium	200.7/6010
Zinc	200.7/6010
Digestions:	
Water	3020
Soil	3050

TABLE 3
Laboratory Equipment and Instrumentation

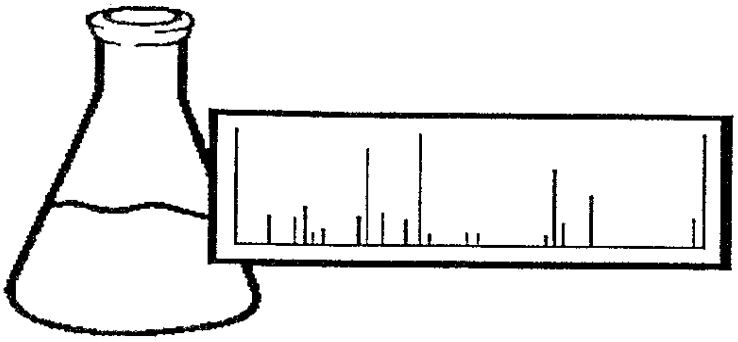
Item	No. of Units	Manufacturer	Model
ANALYTICAL BALANCE 0.1 mg sensitivity,Stable base Class S weights NIST Traceable Service contracts	1	Mettler	AD-160
MAGNETIC STIRRER Variable Speed TFE coated stir bar	3	Corning	PC 351
pH METER ± 0.05 units, Readability ± 0.1 units Line or battery, Usable with specific ion electrodes	1	Orion	940
CONDUCTIVITY METER Readable in ohms or mhos Range of 2 ohms to 2 megaohms, Line or battery	1	YSI	35
HOT PLATE Temperature control	6	Fisher(3) Cimarec(3)	
COLOR STANDARDS To verify wavelengths on photometers Should cover 200 to800 nm	1	Oxford	spectro-check
REFRIGERATOR Sample Storage	3	United	
REFRIGERATOR Standards Storage	4	Avanti, etc.	
DRYING OVEN Gravity or convention, Controlled from room Temperature to 400°C for cleaning organic glass	2	VWR	1305U
THERMOMETERS Mercury-filled celcius, 1°C or finer subdivision to 180°C Certified by or traceable to NIST	15		
GLASSWARE Borosilicate Class A volumetric			

TABLE 3
Laboratory Equipment and Instrumentation

Item	No. of Units	Manufacturer	Model
SPECTROPHOTOMETER	1	Milton Ray	401
Range 400 to 700 nm	1	Bausch & Lomb	700
Band width not greater than 20 nm			
Use several size and shape cells			
Path length 1 to 5 cm			
SPECIFIC ION METER	2	Orion	9401
Readable & accurate to ±1 mV			
ELECTRODES	2	pH, F	960
As needed			
INDUCTIVELY COUPLED PLASMA	1	Leeman	PS1000
Computer control, Background coordination			
Radio frequency generator, Argon gas supply			
WATER BATH	1	Precision	Coliform
Electric or steamed heat	1	Precision	185
Heat to 100°C, Controllable with 5°C			
ION CHROMATOGRAPH	1	Waters	HPLC System
U.V. and Conductivity detector, Separator columns			
	1	Dionex	AS-40/DX-120
Conductivity detector/Carbonate eluent			
ATOMIC ABSORPTION SPECTROPHOTOMETER w/Zeeman Correction	1	Varian	AA-20
Single channel, Single or double beam			
Grating monochrometer, Photomultiplier detector			
Adjustable slits, Range 190 to 800 nm			
Readout system:			
PIPETTORS	8	Finntip/Eppendorf	
Microliter capacity with, disposable tips			
Sizes 5 to 10000 microliters			
RO Water Purification System	1	MilliporeWaters	Milli-Q

TABLE 3
Laboratory Equipment and Instrumentation

Item	No. of Units	Manufacturer	Model
GLASSWARE		Wheaton/Kimble	
Separator Funnels			
Kuderna Danish (K-D), Concentrators			
Water bath for K-D			
GAS CHROMATOGRAPHS	3	Hewelett-Packard	5890
±0.2°C oven, Temperature control	3	Hewelett-Packard	5890 Series II
GC Detectors			
Electron Capture Detector	1	Hewelett Packard	
Flame Ionization Detector	1	Hewelett Packard	
	1	OI Analytical	
Photo-ionization Detector	1	HNU	PI-52
	2	OI Analytical	4560
Mass Spectrometer:	1	Hewelett Packard	5970
	1	Hewelett Packard	5971
GC Purge and trap system	2	Tekmar	ALS/LSC2
	1	Tekmar	LSC2000/ALS2016
	2	OI	MPM-16
GC Semi-Vol. Autosampler	2	Hewelett Packard	7673
Dessiccator	4	Kimax	
Muffle furnace	1	Fisher	184 A
Centrifuge	1	Fisher	Centrific
Fluorometer	1	Turner	20-005 R
Automated Colorimeter	1	Lachat	Quickchem 8000



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RESIDUE, NON-FILTERABLE(TSS)
Method EPA 160.2
Standard Operating Procedure

REVISED: January, 2001

Revision #: 4

Technical Director: _____ **Date :** 3/22/01
Harry B. Locker Ph.D.

QA Officer: _____ **Date :** 3/22/01
Mark L. Fausel

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I. Overview:

-Method Summary - to determine the amount of non-filterable residue based on a gravimetric determination. The sample matrix is generally aqueous.

II. Safety: Health = 1 Fire = 0 Reactivity = 0

-All personal protective equipment, (gloves, goggles and lab coat), should be worn when handling any samples and reagents.
-Samples are filtered through Gooch crucibles at the filter box.
-There are no special health risks associated with this method.

III. Waste Disposal:

-Sample that passes through the filter paper and collects in the filter box is dumped down the drain.
-Paper filters are scraped out of each crucible and are put into the solid waste satellite container.
-Crucible are soaked in 1:1 HCl, washed and rinsed before reuse..

IV. Sample Collection, Preservation, Storage:

-Samples may be collected in plastic jugs or glass jars.
-Generally, samples may be stored on the shelving located under the chemical cabinets in the inorganic lab or in the inorganic lab sample refrigerator.
-There is no sample preservation.
-Total suspended solids have a holding time of 7 days.

V. Equipment & Supplies:

-Filter Box
-Glass Fiber Filters, (Whatman 934-AH, 21 or 24 mm diameter, 1.5 µm pore size)
-Gooch Crucibles
-Drying Oven
-Desiccator
-Balance
-Crucible Adapter

VI. Reagents & Standards:

-AlphaTrol solution is used as the laboratory fortified blank. None of this solution is added to any of the actual samples.

VII. Interferences:

-Interferences may occur from leftover residue in crucibles from prior analysis.

-Large floating particles or non-homogeneous matter should be excluded from the sample.
-Variations in cooling periods between weightings can alter the sample weights.

VIII. Quality Control:

-IDC - All analysts of this method must perform an IDC that meets standard requirements.
-Laboratory Reagent Blanks are run 1 per set.
-Laboratory Fortified Blanks with a target of 80 ppm, are run 2 per set.
-No lab Fortified Matrix Samples are performed by this analysis.
-All samples are analyzed in duplicate.
-Control Charts - Each analyst should familiarize themselves with the control charts for this analysis. This chart will tell you the acceptance values, warning limits and control limits for LFB's and duplicates.
-This method has a reporting limit of 2 mg/L.

IX. Analytical Procedure:

-Prepare glass fiber filter by placing it rough side up in bottom of Gooch crucible.
    -There are 2 sizes of crucible: 24cm and 21cm.
    -Crucibles must have the proper size filter for proper results.
-Using the crucible adapters and filter box, pass DI water through filter three times in 20 mL increments. Dry crucible in vented oven at 103-105°C for one hour. Cool, desiccate for at least two hours, and weigh.
-Gather all samples. Shake samples well and measure an appropriate amount of sample (100 mls, 50 mls, or 30 mls based on the nature of the sample) with a graduated cylinder. Samples may be diluted if necessary.
-Filter the sample through the crucible until all traces of water are removed by continuing vacuum after all of the sample has passed through the filter and crucible. Rinse graduated cylinder 3 times between samples.
-If more sample has been put into the crucible than will filter through, that crucible must be scrapped and a fresh one used.
-Dry crucible in oven at 103-105°C for at least two hours. Cool, desiccate at least two hours and weigh.
-Dry for another 1-2 hours, cool in desiccator for at least two hours and reweigh. Repeat until difference in weighings are 0.0005g or less.

X. Calibration & Standardization:

-Calibration of scale only. See "Scale Maintenance" in "General Laboratory Log Book".

XI. Data Reduction:

Total Suspended Solids

$$mg/L = \frac{(A-B) \times 1000}{C}$$

A = mass of crucible and residue in grams
B = mass of crucible in grams
C = liters of sample filtered

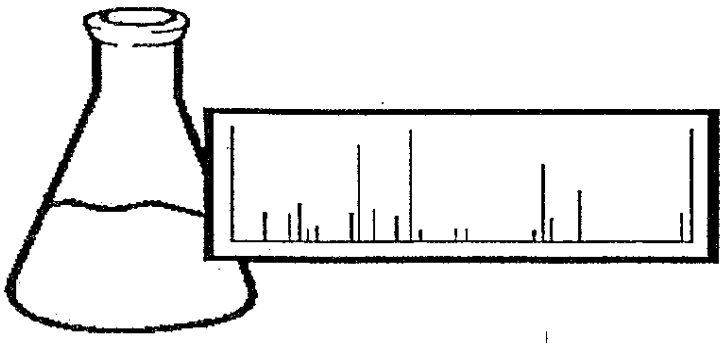
XII. Data Recording:

-Data should be entered directly into the excel benchsheet [ezekial/excel/nutrient/tss/tsstemp.xlt].
-A printoout of the benchsheet should be placed into the solids binder.
-Final results should be recorded in the LIMS with analysts initials, date analyzed, and units.
-Results < 10 ppm should be recorded with 1 significant figure, but not past the decimal point; 10-99 ppm should be recorded with 2 significant figures, but not past the decimal point; > 100 ppm should be recorded with 3 significant figures, but not past the decimal point.
-Soft data is archived monthly on a zip disk labeled "General Inorganics Data".
-For long term storage, hard data is stored annually in a bankers box labeled "General Inorganics Data".

XIII. Troubleshooting:

-Make sure that the duration of time the crucibles spend in the dessicator between weighings is constant.
-If weighings are greater than 0.0005g apart, remove crucible from balance, dust off inside of balance, retare and reweigh crucible. If the variation remains, then reheat and the samples for a third time.
-If balance continually adjusts, desiccant in desiccators may need reconditioning, or the scale may need recalibration.

XIV. Method Modifications:



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TOTAL PHOSPHOROUS/TOTAL DISSOLVED PHOSPHOROUS
Method SM 4500-p F/EPA 365.1
Standard Operating Procedure

REVISED: January, 2001

Revision #: 3

Technical Director: _____ **Date :** 3/27/01
Harry B. Locker Ph.D.

QA Officer: _____ **Date :** 3/27/01
Mark L. Faysel

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I. Overview:

-This method covers the determination of total phosphorus in drinking, ground and surface water, and domestic and industrial wastes. This method determines total phosphorus, or if the sample is filtered through a 0.45 micron pore size filter, the result is termed total dissolved phosphorus. The difference between the result of a sample determined directly and filtered is termed total insoluble phosphorus. A sample that is analyzed without preliminary digestion is termed orthophosphorus. This method also includes available phosphorus and reserve phosphorus.
-The orthophosphate ion (PO_4^{3-}) reacts with ammonium molybdate and antimony potassium tartrate under acidic conditions to form a complex. This complex is reduced with ascorbic acid to form a blue complex which absorbs light at 880 nm. The absorbance is proportional to the concentration of orthophosphate in the sample.

II. Safety:

A. Wear safety glasses, gloves and lab coat when handling any reagents or chemicals.
B. During digestion, small amounts of sulfuric acid fumes and aerosol are released. Therefore, all digestions must take place in a ventilated fume hood.
C. The following chemicals should be handled with extreme caution:
    1. Sulfuric acid - reacts violently with water; extremely corrosive; will cause extreme burns on skin; use in a fume hood
    2. Antimony Potassium Tartrate - NFPA Health Rating of 3. Use of a dust mask is recommended.

III. Waste Disposal:

-The waste stream from the Lachat is collected in a benchtop carboy. It is then disposed of by flushing down the sink with copious amounts of water.

IV. Sample Collection, Preservation and Storage:

-Samples should be collected in glass bottles. Plastic bottles are not acceptable because low concentration of phosphates may be absorbed onto the walls of plastic bottles.
-Preservation Methods are as follows:
    -Total Phosphorus
        -Cool, 4°C
        -H_2SO_4 to pH <2
    -Total Dissolved Phosphorus
        -Filter
        -Cool, 4°C
        -H_2SO_4 to pH <2
    -Orthophosphate
        -Filter

-Cool, 4°C

-Acid preserved samples have a holding time of 28 days. Samples not preserved with acid have a holding time of 48 hours. Samples are stored in the Inorganic Refrigerator.

V. Equipment and Supplies:

-Flow injection analysis equipment, including autosampler, pump, reaction manifold, colorimetric detector and data system.
-BD-46 Block Digestor
-Vortex Mixer
-Polystyrene Culture Tubes
-75 ml glass digestion tubes with stainless steel rack
-Hengar Boiling Chips (Selenized chips are acceptable)

VI. Reagents and Standards:

A. Reagents

Reagent 1. Stock Ammonium Molybdate Solution

-In a 1 L volumetric flask dissolve **40.0 g ammonium molybdate tetrahydrate** $[(NH_4)_6Mo_7O_{24}4H_2O]$ in approximately **800 mL** water. Dilute to the mark and invert to mix. STIR AT LEAST 4 HOURS. STORE IN PLASTIC AND REFRIGERATE.

Reagent 2. Stock Antimony Potassium Tartrate Solution

-In a 1 L volumetric flask, dissolve **3.0 g antimony potassium tartrate** (potassium antimony tartrate hemihydrate $k(SbO)C_4H_4O_6$ $1/2H_2O$) in approximately **800 mL** of water. Dilute to the mark and invert to mix. Store in a dark plastic bottle and refrigerate.

Reagent 3. Molybdate Color Reagent

-To a 1 L volumetric flask add about **500 mL** water, and then add **21.0 mL concentrated sulfuric acid** (CAUTION: The solution will get very hot!) Swirl to mix. When it can be comfortably handled, add **72.0 mL Stock Antimony Potassium Tartrate Solution** (Reagent 2) and **213 mL Ammonium Molybdate Solution** (Reagent 1). Dilute to the mark and invert to mix. Store in dark glass bottle. Allow several hours for reagent to degas.

Reagent 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** of water. Dilute to the mark and invert to mix. Add **1.0 g dodecyl sulfate** $[CH_3(CH_2)_{11}OSO_3Na]$. **Prepare fresh weekly. Discard if the solution becomes yellow.**

Reagent 5. Carrier, 0.13M Sulfuric Acid

-In a 1 L volumetric flask add 500 ml water and 7.2 ml concentrated sulfuric acid (H_2SO_4). Dilute to the mark and invert to mix. Prepare fresh weekly.

Reagent 6. Diluent

-The diluent is the same as the carrier (above). It is used to dilute digested samples.

B. Standards shall be prepared from LabChem Phosphate (P) Standard, 50 ppm (1 ml = 0.05 mg P), ranging from 0 ppb to 1000 ppb. A suggested set is as follows:

Made up in 20.0 mls DI
-0 ppb
-5 ppb (20 ul of 5 ppm)
-12.5 ppb (50 ul of 5 ppm)
-25 ppb (100 ul of 5 ppm)
-50 ppb (200 ul of 5 ppm)
-100 ppb (400 ul of 5 ppm)
-400 ppb (160 ul of 50 ppm)
-800 ppb (320 ul of 50 ppm)
-1000 ppb (400 ul of 50 ppm)

VII. Interferences:

1. Glassware contamination is a problem in low level phosphorus determinations. Glassware should be washed with phosphate - free detergent, soaked in 1:1 HCl, and rinsed with deionized water.
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. Samples high in iron can be pretreated with Sodium Bisulfate to eliminate this interference.
3. Contamination of total dissolved phosphorus samples may be controlled by acid rinsing the filter funnels with 10% H_2SO_4.

VIII. Quality Control:

A. Initial Demonstration of Capability (DOC) - Each analyst must demonstrate method proficiency through an DOC. A series of 4 mid-level LFB samples are prepared and analyzed. Standard recovery must fall within ±20% .
B. The following solutions should be analyzed every 10 samples:
-Lab Reagent Blank - must fall below reporting limit
-Lab Fortified Blank - ±20% Recovery
C. The following solutions should be analyzed every 20 samples:
-Lab Fortified Sample Matrix
-Lab Duplicate
D. Each analyst will document Quality Control information on a "Weekly Analyst QC Table". These tables are submitted to the Laboratory QC Officer. Quarterly Control Charts are generated, and will present upper and lower method control limits.
E. QC and sample data are stored in a 3-ring binder labeled "Lachat Data" in the Inorganic Department.

IX. Analytical Procedure:

A. Sample Preparation

1. Filtration

a. Total Dissolved Phosphorus - filter through a 0.45 µ filter

b. Orthophosphate - filter through a 0.45 µ filter. DO NOT DIGEST (proceed directly to Data Acquisition).

c. Available and Reserve Phosphorus -- See Appendix A

2. Digestion

a. Samples are digested in 75 ml glass digestion tubes. To each digestion tube add:

1. 20.0 ml sample or standard

*NOTE: High concentration samples should be diluted prior to digestion. Suggested dilutions are as follows:

Dairy Waste	1:100
Treatment Plant Influents	1:5
Treatment Plant Effluents	1:2 or 1:5

2. Pinch Potassium Persulfate ($K_2S_2O_8$) (Approximately 0.32g)
3. 400 ul 11 N Sulfuric Acid
4. 2 Hengar Boiling Chips

-Rinse chips with 1:1 HCl, then DI.

b. Spray the outside of the tubes generously with Plasti-Kote Silicone Spray. CAREFULLY AND SLOWLY lower the sample tubes into the digester so that the rack rests on the block housing.

c. Digest for 30 minutes at 160°C. Carefully remove the hot tubes from the digester and place in the rack base. Allow the samples to cool and then add 8-10 mls of DI.

d. Vortex each digestion tube to thoroughly mix the sample. Transfer to a clean, acid rinsed 50 ml graduated plastic cup. Bring the sample volume up to 20.0 ml with DI.

e. Transfer an aliquot of the sample into a polystyrene culture tube and place in the appropriate spot in the autosampler rack.

B. Data Acquisition

1. Turn on system and log into Omnion. Remember the system boots up to the last files that were open.
2. Create your new method. *File, New Method* to define Analyte Table. Define *Method, Valve Timing, Sampler Timing*, and *Pump Timing*. Save defined method. *File, Save method As...* or Open the method you want to run. *file, open method....*
3. Create your new tray. *File, New Tray*. Save defined tray. *File, save tray as.* or To run the same samples as an earlier tray, file open tray.
4. Create your own new data quality management plan. *File, new DQM plan.* Save defined data quality management plan. *File, save DQM Plan* as... or to use a saved DQM plan, file, *open DQM plan....*
5. Allow heating unit to heat to 37°C.
6. Install the manifold.

7. Turn on pump. Snap pump tube cartridges into position.
8. Place reagent transmission lines into DI.

9. Pump DI into manifold and check for possible leaks. Place reagent transmission lines into the appropriate containers. Allow reagents to stabilize.
10. Run the analysis. *Tray, Run Tray.* While tray is processing, you can view the peaks. *Window, channel name window. Data, split screen*, and click and drag desired areas for different views. You will see the message *tray run complete* when the *STOP* tuns to *run tray.*
11. Update analyte table with defined graphical events (if necessary). View peak integration window. *Window, channel name window. Method, graphical events programming, peak base width and threshold.* Save any changes made to analyte table. *File, save method.*
12. Graphically determine the proper inject to peak start. View peak integration window. *Window, channel name window.* Graphically define the *peak start* and *peak stop.*
13. Reanalyze the data using updated method (if necessary). *Data, reanalyze data.*
14. View the calibration. *Method, review analyte calibration curve.*
15. Prepare a customized report for your results. *Method, custom report.*

NOTE: Steps 11-13 are not always necessary.

C. Data system parameters for Quikchem 8000

-The timing values listed below are approximate and will need to be optimized using graphical events programming.

Sample Throughput:	80 samples/h, 45 s/sample
Pump Speed:	35
Cycle Period:	35

Analyte Data:

Concentration Units:	ug P/L
Peak Base Width:	36 s
% Width Tolerance:	100
Threshold:	2500
Inject to Peak Start:	11 s
Chemistry:	Direct

Calibration Data:

Level	Concentration (ug/L)
1	0
2	5
3	12.5
4	25
5	50
6	100
7	400
8	800

9 1000

Calibration Fit Type:	1st Order Polynomial
Calibration Rep. Handling:	Weighted Average
Weighing Method:	1/X
Concentration Scaling:	None
Force Through Zero:	No

Sampler Timing:

Min. Probe in Wash Period:	10 s
Probe in Sample Period:	23 s

Valve Timing:

Load Time:	0.0 s
Load Period:	18 s
Inject Period:	27 s

D. System Shut Down

1. Remove the reagent lines from each reagent and rinse off the lines and glass weights before putting them in DI rinse.
2. Allow DI to flush through the system for 5-10 minutes at "normal" speed.
3. Remove the transmission lines from the DI and allow all liquid to be pumped out of the manifold.
4. Turn off the pump and release the tension on the pump tube cartridge. DO NOT LEAVE ANY PUMP TUBES CLAMPED DOWN WHEN THE PUMP IS SHUT OFF.
5. Disconnect the manifold from the Quickchem 8000. Carefully wind the transmission lines around the manifold for storage.

X. Generation of Calibration Line:

-Prepare standards in DI and digest them according to step IX.A.2. Calibration is performed by injecting the standards. The data system will then prepare a calibration curve by plotting response versus standard concentration. Sample concentration is calculated from the regression equation.
-It is recommended that a fresh calibration line is digested and analyzed each analytical day (especially) if fresh reagents have been prepared since the last calibration was performed. If this is not possible, a line check must be analyzed to verify the integrity of the line. The line check must fall within ±20%.

XI. Data Reduction:

-Remember that Lachat results are presented as µg/L. Convert µg/L readings to mg/L.
-Insure that any dilutions are factored into the final result.
-Calculate QC and Spike recoveries, and the difference between Repeat Analyses.

XII. Data Recording:

A. Samples

1. The following is a flow chart for Inorganic samples throughout the laboratory:

a. Samples enter the lab, are logged in and are assigned a reference number.

b. The chain of custody report is placed in the incoming basket in the Inorganic Department.

c. The analyst may create a "worklist" using the sample master LIMS to determine which samples to analyze.

d. All analyses are recorded in the digestion log labeled "TP/TDP Digestion Log". Each log sheet should include the sample reference number, analysts' name, LFB concentrations and lot number, LFM concentrations and lot number and a listing of the reagents used including lot number.

e. Raw data is stored in a binder labeled "Lachat Data" located in the Inorganic Department.

f. For long term storage, data is stored in a bankers box labeled "Lachat Raw Data".

XIII. Instrument Maintenance:

A. BD-46 Block Digeter-(see BD-46 Block Digester User's Guide, p. 18-19).

B. Quickchem 8000-Keep all modules clean and dry at all times.

C. Autosampler

1. It may be necessary to use a light machine oil on the x-axis shaft, y-axis shaft, and the four idler pulley shafts, on a periodic basis depending on the laboratory environment.

2. Ensure that the probe is straight and not damaged in any way.

D. Watson-Marlow Pump

1. The pump is very susceptible to acid damage. Immediately wipe off any spills.

2. The rollers of the pump may get rusty and can be cleaned with steel wool. A very light coat of silicone spray can be applied by spraying it on a cloth and holding it on the moving roller. The silicone will act as a rust inhibitor.

3. Check for cracks or acid damage on the cartridges.

E. Valve Modules

1. Valve modules may be damaged by over-tightening the valve fittings or by liquid spills.

a. Replace the o-rings once per month.

b. When changing o-rings, clean the valve ports with a cotton swab and DI.

XIV. Troubleshooting:

A. If a blue color appears in the tubing, or if the baseline drift upward, place the reagent transmission lines into a NaOH-EDTA solution (65 g NaOH + 6 g disodium EDTA in 1 L of DI water). Pump this solution for 5 minutes.
B. If the Ascorbic Acid Reducing Solution (Reagent #4) has a lot of air bubbles in it, allow the solution to settle for several hours before use.
C. Record all troubleshooting information in the "Lachat Troubleshooting" binder, stored on the Lachat workbench in the Inorganic Department.

XV. Method Modifications:

APPENDIX A: AVAILABLE P AND RESERVE P

A. Equipment:

1. Rotary Shaker, capable of 180 oscillations per minute
2. 125 ml Erlenmeyer flasks

B. Reagents:

1. Modified Morgan Estrextant
Add 71.9 ml glacial acetic acid to approx. 500 ml DI in a 1 L volumetric flask. Add 45.6 ml concentrated NH_4OH. Dilute to 1 L and mix well. Adjust the pH to $4.8 \pm .05$ with either concentrated NH_4OH or acetic acid.

C. Procedure:

1. Place 10.0 g air-dried, sieved soil in a 125 ml Erlenmeyer flask.
2. Add 50 ml extractant.
3. Shake at 180 oscillations per minute for 15 minutes on a reciprocating shaker.
4. Filter through a medium porosity filter paper (Whatman No. 2 or equivalent).
5. Analyze filtrate for P.

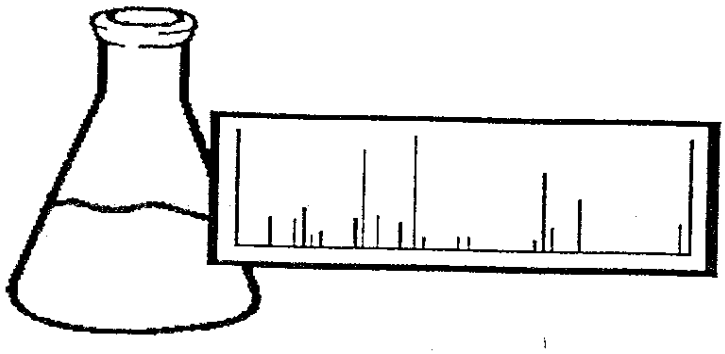
-Proceed to step IX.B. Data Acquisition

D. Data Reduction:

1. Report Available P as mg/kg dry weight.

$$\frac{(\text{mg/L})\ (\text{Final vol. in L})}{(\%\ \text{solid as a decimal})\ (\text{mass in grams})} \times 1000\ \text{g/kg} = \text{mg/kg dry weight}$$

2. (Total Phosphorus) - (Available Phosphorus) = Reserve Phosphorus



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ESCHERICHIA COLI
Method SM 9223B
Standard Operating Procedure

REVISED: January, 2001

Revision #: 2

Technical Director: _____ Date : 3/27/01
Harry B. Locker Ph.D.

QA Officer: _____ Date : 3/27/01
Mark L. Fausel

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I. Overview:

Colisure is used for the simultaneous detection and confirmation of total coliforms and E.coli in fresh and marine waters. When total coliforms metabolize Colisure's nutrient, ONPG, the sample turns purple. When E.coli metabolize Colisure's nutrient indicator, MUG, the sample fluoresces. Colisure can simultaneously detect these bacteria at 1 MPN/100ml within 24-48 hours even with as many as 2 million heterotrophic bacteria per 100ml present.

II. Safety:

Wear safety glasses, gloves, and lab coat when handling media or samples.

III. Waste Disposal:

Negative samples may be disposed of in the trash. Positive samples must be autoclaved for 30 minutes at 124°C before disposal.

IV. Sample Collection, Preservation, and Storage:

Holding time-6 hours at 4°C
Sample storage-samples are stored in the Inorganics refrigerator

V. Equipment and Supplies:

1. IDEXX Colisure granulated media, catalog #WP200 (store at 4-25°C away from light)
2. IDEXX Quanti-Tray or Quanti-Tray 2000
3. IDEXX Quanti-Tray Sealer
4. Autoclave (Napco 8000 DSE)
5. Fisher 146 low temperature incubator
6. Stock solutions of E.coli, P aeruginosa, K. pneumoniae
7. 6 watt, 365nm UV light
8. Prefilled phosphate A 99ml sterile diluent bottles, Fisher.
9. IDEXX well quanti-tray MPN table 2000 or 51

VI. Reagents and Standards:

NA

VII. Interferences:

NA

VIII. Quality Control:

A. The following quality control procedure is recommended for each lot of Colisure:
   1. Inoculate 3 sterile vessels filled with 100ml sterile water with the following:
      a. One with a sterile loop of E.coli ATCC*25922
      b. One with a sterile loop of Klebsiella pneumoniae ATCC31488
      c. One with a sterile loop of Pseudomonas aeruginosa ATCC10145
   2. Follow the P/A procedure or Quanit-Tray Enumeration Procedure
   3. Results should match the result interpretation table in section IX

B. Prepare one positive and one negative control for each analytical day. The positive control is prepared by inoculating sterile DI with a loopful of E.coli. The negative control is prepared by inoculating sterile DI with P. aeruginosa.

IX. Analytical Procedure:

A. Quanti-Tray Enumeration Procedure
   1. Add contents of one snap pack to a 100ml, room temperature water sample in a sterile vessel. Vessel does not need to be transparent or nonfluorescing
   2. Cap vessel and shake until dissolved
   3. Pour sample/reagent mixture into a Quanti-Tray or Quanti-Tray/2000 and seal in an IDEXX Quanti-Tray Sealer
   4. Place the sealed tray in a 35±0.5°C incubator for 24-48 hours
   5. Read results according to the result interpretation table below. Count the number of positive wells and refer to the MPN table to obtain a most probable number

      Note: If using the Quanti-Tray/2000, tap the tray to remove air bubbles from the wells

B. Result Interpretation

Appearance	Result
Yellow or slight tinge	negative for total coliform and e.coli
Purple equal to or greater than the comparator	positive for total coliform
Purple and fluorescence equal to or greater than the comparator	positive for e.coli

   1. Look for fluorescence with a 6 watt, 365nm, UV light within 5 inches of the sample. Face light away from your eyes and towards the sample. Wear UV protective eyeware
   2. Samples are negative if at any time after 24 hours there is no purple and/or fluorescence
   3. Purple or purple/fluorescence observed before 24 hours is a valid positive. However, after 48 hours from inoculation, heterotrophs may overwhelm Colisure's inhibition system. Therefore, purple or purple/fluorescence first observed after 48 hours from inoculation is not a valid positive

C. Procedural Notes
   1. A slight tinge may be observed when Colisure is added to the sample

2. Some water samples containing humic material may have an innate color. If a water sample has some background color, compare inoculated Colisure sample to a control blank of the same water sample
3. Do not use buffered water in this procedure. Colisure media is buffered

X. Calibration and Standardization:

NA

XI. Data Reduction:

NA

XII. Data Recording:

A. Enumeration Procedure
1. Use number count and translate to MPN/100ml with use of IDEXX 2000 MPN table. Final results are entered and saved in LIMS.

XIII. Instrument Maintenance:

Ice build up in the incubator requires periodic defrosting. If incubator temperature does not remain stable or continues to drop, defrost the incubator.

XIV. Trouble Shooting:

XV. Method Modifications:

* American Type Culture Collection 1-800-638-6597