PhycoTech, Inc.
620 Broad Street - Suite 100 - St. Joseph - MI 49085 - Phone: 269-983-3654 - Fax: 269-983-3653
info@phycotech.com - www.phycotech.com

August 25, 2008

WV Purchasing Division
2019 Washington St E
PO Box 50130
Charleston WV 25305-0130

Mr. Chuck Bowman,

Attached please find our response to your RFQ Number DEP13876 for the Processing and Identification of Diatoms & Soft Algae from Periphyton Samples.

The pricing we have provided would be based on our desire to do the QA/QC portion of the analysis only as referenced in the scope of work. We would be able to process a total of 70 samples per year of QA/QC data.

We have attached for your review the completed and signed RFQ document, the resume of Ann St. Amand, PhycoTech, Inc. Quality Assurance Plan, PhycoTech, Inc. General Technical Approach and a sample data report.

If you need additional information after reviewing these items please visit our website at www.phycotech.com or contact us directly.

Thank you for this opportunity.

Sincerely.

Ann St. Amand
President

State of West Virginia
Department of Administration
Purchasing Division
2019 Washington Street East
Post Office Box 50130
Charleston, WV 25305-0130

Request for Quotation

RFQ NUMBER	PAGE
DEP13876	1

ADDRESS CORRESPONDENCE TO ATTENTION OF:
CHUCK BOWMAN
304-558-2157

VENDOR
PhycoTech, Inc.
620 Broad Street
Suite 100
St. Joseph, MI 49085

SHIP TO
ENVIRONMENTAL PROTECTION
DEPARTMENT OF
OFFICE OF WATER RESOURCES
601 57TH STREET SE
CHARLESTON, WV
25304 304-926-0499

DATE PRINTED	TERMS OF SALE	SHIP VIA	F.O.B.	FREIGHT TERMS
07/31/2008				

BID OPENING DATE: 08/28/2008 BID OPENING TIME 01:30PM

LINE	QUANTITY	UOP	CAT. NO.	ITEM NUMBER	UNIT PRICE	AMOUNT
0001	1	JB		493-09		

WATER, WASTE WATER AND SOIL SAMPLE ANALYSIS

OPEN END CONTRACT

THE WEST VIRGINIA PURCHASING DIVISION, FOR THE AGENCY, THE WEST VIRGINIA DEPARTMENT OF ENVIRONMENTAL PROTECTION, IS SOLICITING BIDS TO PROVIDE FOR THE PROCESSING AND IDENTIFICATION OF PERIPHYTON SAMPLES FOR THE DIVISION OF WATER AND WASTE MANAGEMENT, WATERSHED ASSESSMENT BRANCH, PER THE ATTACHED SPECIFICATIONS, TERMS, CONDITIONS AND BID REQUIREMENTS.

PLEASE NOTE THERE ARE MANDATORY BID SUBMISSION REQUIR-MENTS LISTED UNDER "QUALIFICATIONS" IN THE ATTACHED SPECIFICATIONS. THE QUALIFICATION CRITERIA LISTED IS MANDATORY AND MUST ACCOMPANY THE BID SUBMITTAL IN ORDER FOR THE BID TO BE CONSIDERED FOR AWARD. FAILURE TO INCLUDE THIS INFORMATIN WILL RESULT IN DISQUALIFI-CATION OF THE VENDOR'S BID.

EXHIBIT 3

LIFE OF CONTRACT: THIS CONTRACT BECOMES EFFECTIVE UPON THE AWARD AND EXTENDS FOR A PERIOD OF ONE (1) YEAR OR UNTIL SUCH "REASONABLE TIME" THEREAFTER AS IS NECESSARY TO OBTAIN A NEW CONTRACT OR RENEW THE ORIGINAL CONTRACT. THE "REASONABLE TIME" PERIOD SHALL NOT EXCEED TWELVE (12) MONTHS. DURING THIS "REASONABLE TIME" THE VENDOR MAY TERMINATE THIS CONTRACT FOR ANY REASON UPON GIVING THE DIRECTOR OF PURCHASING 30 DAYS

SEE REVERSE SIDE FOR TERMS AND CONDITIONS

SIGNATURE	TELEPHONE	DATE

TITLE	FEIN	ADDRESS CHANGES TO BE NOTED ABOVE

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GENERAL TERMS & CONDITIONS
REQUEST FOR QUOTATION (RFQ) AND REQUEST FOR PROPOSAL (RFP)

1. Awards will be made in the best interest of the State of West Virginia.

2. The State may accept or reject in part, or in whole, any bid.

3. All quotations are governed by the ***West Virginia Code*** and the ***Legislative Rules*** of the Purchasing Division.

4. Prior to any award, the apparent successful vendor must be properly registered with the Purchasing Division and have paid the required $125.00 registration fee.

5. All services performed or goods delivered under State Purchase Orders/Contracts are to be continued for the term of the Purchase Order/Contract, contingent upon funds being appropriated by the Legislature or otherwise being made available. In the event funds are not appropriated or otherwise available for these services or goods, this Purchase Order/Contract becomes void and of no effect after June 30.

6. Payment may only be made after the delivery and acceptance of goods or services.

7. Interest may be paid for late payment in accordance with the ***West Virginia Code.***

8. Vendor preference will be granted upon written request in accordance with the ***West Virginia Code.***

9. The State of West Virginia is exempt from federal and state taxes and will not pay or reimburse such taxes.

10. The Director of Purchasing may cancel any Purchase Order/Contract upon 30 days written notice to the seller.

11. The laws of the State of West Virginia and the ***Legislative Rules*** of the Purchasing Division shall govern all rights and duties under the Contract, including without limitation the validity of this Purchase Order/Contract.

12. Any reference to automatic renewal is hereby deleted. The Contract may be renewed only upon mutual written agreement of the parties.

13. **BANKRUPTCY:** In the event the vendor/contractor files for bankruptcy protection, this Contract may be deemed null and void, and terminated without further order.

14. **HIPAA Business Associate Addendum:** The West Virginia State Government HIPAA Business Associate Addendum (BAA), approved by the Attorney General, and available online at the Purchasing Division's web site **(http://www.state.wv.us/admin/purchase/vrc/hipaa.htm)** is hereby made part of the agreement. Provided that, the Agency meets the definition of a Covered Entity (45 CFR §160.103) and will be disclosing Protected Health Information (45 CFR §160.103) to the vendor.

15. **West Virginia Alcohol & Drug-Free Workplace Act:** If this Contract constitutes a public improvement construction contract as set forth in Article 1D, Chapter 21 of the West Virginia Code ("The West Virginia Alcohol and Drug-Free Workplace Act"), then the following language shall hereby become part of this Contract: "The contractor and its subcontractors shall implement and maintain a written drug-free workplace policy in compliance with the West Virginia Alcohol and Drug-Free Workplace Act, as set forth in Article 1D, Chapter 21 of the West Virginia Code. The contractor and its subcontractors shall provide a sworn statement in writing, under the penalties of perjury, that they maintain a valid drug-free work place policy in compliance with the West Virginia Alcohol and Drug-Free Workplace Act. It is understood and agreed that this Contract shall be cancelled by the awarding authority if the Contractor: 1) Fails to implement its drug-free workplace policy; 2) Fails to provide information regarding implementation of the contractor's drug-free workplace policy at the request of the public authority; or 3) Provides to the public authority false information regarding the contractor's drug-free workplace policy."

INSTRUCTIONS TO BIDDERS

1. Use the quotation forms provided by the Purchasing Division.

2. **SPECIFICATIONS:** Items offered must be in compliance with the specifications. Any deviation from the specifications must be clearly indicated by the bidder. Alternates offered by the bidder as **EQUAL** to the specifications must be clearly defined. A bidder offering an alternate should attach complete specifications and literature to the bid. The Purchasing Division may waive minor deviations to specifications.

3. Complete all sections of the quotation form.

4. Unit prices shall prevail in cases of discrepancy.

5. All quotations are considered F.O.B. destination unless alternate shipping terms are clearly identified in the quotation.

6. **BID SUBMISSION:** All quotations must be delivered by the bidder to the office listed below prior to the date and time of the bid opening. Failure of the bidder to deliver the quotations on time will result in bid disqualifications:
Department of Administration, Purchasing Division, 2019 Washington Street East, P.O. Box 50130, Charleston, WV 25305-0130

State of West Virginia
Department of Administration
Purchasing Division
2019 Washington Street East
Post Office Box 50130
Charleston, WV 25305-0130

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VENDOR
RFQ COPY
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WRITTEN NOTICE.

UNLESS SPECIFIC PROVISIONS ARE STIPULATED ELSEWHERE IN THIS CONTRACT DOCUMENT, THE TERMS, CONDITIONS AND PRICING SET HEREIN ARE FIRM FOR THE LIFE OF THE CONTRACT.

RENEWAL: THIS CONTRACT MAY BE RENEWED UPON THE MUTUAL WRITTEN CONSENT OF THE SPENDING UNIT AND VENDOR, SUBMITTED TO THE DIRECTOR OF PURCHASING THIRTY (30) DAYS PRIOR TO THE EXPIRATION DATE. SUCH RENEWAL SHALL BE IN ACCORDANCE WITH THE TERMS AND CONDITIONS OF THE ORIGINAL CONTRACT AND SHALL BE LIMITED TO TWO (2) ONE (1) YEAR PERIODS.

CANCELLATION: THE DIRECTOR OF PURCHASING RESERVES THE RIGHT TO CANCEL THIS CONTRACT IMMEDIATELY UPON WRITTEN NOTICE TO THE VENDOR IF THE COMMODITIES AND/OR SERVICE SUPPLIED ARE OF AN INFERIOR QUALITY OR DO NOT CONFORM TO THE SPECIFICATIONS OF THE BID AND CONTRACT HEREIN.

OPEN MARKET CLAUSE: THE DIRECTOR OF PURCHASING MAY AUTHORIZE A SPENDING UNIT TO PURCHASE ON THE OPEN MARKET, WITHOUT THE FILING OF A REQUISITION OR COST ESTIMATE, ITEMS SPECIFIED ON THIS CONTRACT FOR IMMEDIATE DELIVERY IN EMERGENCIES DUE TO UNFORESEEN CAUSES (INCLUDING BUT NOT LIMITED TO DELAYS IN TRANSPORTATION OR AN UNANTICIPATED INCREASE IN THE VOLUME OF WORK.)

QUANTITIES: QUANTITIES LISTED IN THE REQUISITION ARE APPROXIMATIONS ONLY, BASED ON ESTIMATES SUPPLIED BY THE STATE SPENDING UNIT. IT IS UNDERSTOOD AND AGREED THAT THE CONTRACT SHALL COVER THE QUANTITIES ACTUALLY ORDERED FOR DELIVERY DURING THE TERM OF THE CONTRACT, WHETHER MORE OR LESS THAN THE QUANTITIES SHOWN.

SEE REVERSE SIDE FOR TERMS AND CONDITIONS

SIGNATURE	TELEPHONE	DATE

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ORDERING PROCEDURE: SPENDING UNIT(S) SHALL ISSUE A WRITTEN STATE CONTRACT ORDER (FORM NUMBER WV-39) TO THE VENDOR FOR COMMODITIES COVERED BY THIS CONTRACT. THE ORIGINAL COPY OF THE WV-39 SHALL BE MAILED TO THE VENDOR AS AUTHORIZATION FOR SHIPMENT, A SECOND COPY MAILED TO THE PURCHASING DIVISION, AND A THIRD COPY RETAINED BY THE SPENDING UNIT.

BANKRUPTCY: IN THE EVENT THE VENDOR/CONTRACTOR FILES FOR BANKRUPTCY PROTECTION, THIS CONTRACT IS AUTOMATICALLY NULL AND VOID, AND IS TERMINATED WITHOUT FURTHER ORDER.

THE TERMS AND CONDITIONS CONTAINED IN THIS CONTRACT SHALL SUPERSEDE ANY AND ALL SUBSEQUENT TERMS AND CONDITIONS WHICH MAY APPEAR ON ANY ATTACHED PRINTED DOCUMENTS SUCH AS PRICE LISTS, ORDER FORMS, SALES AGREEMENTS OR MAINTENANCE AGREEMENTS, INCLUDING ANY ELECTRONIC MEDIUM SUCH AS CD-ROM.

REV. 04/11/2001

EXHIBIT 10

REQUISITION NO.: .....

ADDENDUM ACKNOWLEDGEMENT

I HEREBY ACKNOWLEDGE RECEIPT OF THE FOLLOWING CHECKED ADDENDUM(S) AND HAVE MADE THE NECESSARY REVISIONS TO MY PROPOSAL, PLANS AND/OR SPECIFICATION, ETC.

ADDENDUM NO.'S:

SEE REVERSE SIDE FOR TERMS AND CONDITIONS

SIGNATURE	TELEPHONE	DATE

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Request for Quotation

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VENDOR
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NO. 1 .....

NO. 2 .....

NO. 3 .....

NO. 4 .....

NO. 5 .....

I UNDERSTAND THAT FAILURE TO CONFIRM THE RECEIPT OF THE ADDENDUM(S) MAY BE CAUSE FOR REJECTION OF BIDS.

VENDOR PREFERENCE CERTIFICATE

CERTIFICATION AND APPLICATION* IS HEREBY MADE FOR PREFERENCE IN ACCORDANCE WITH WEST VIRGINIA CODE, 5A-3-37 (DOES NOT APPLY TO CONSTRUCTION CONTRACTS).

A. APPLICATION IS MADE FOR 2.5% PREFERENCE FOR THE REASON CHECKED:

( ) BIDDER IS AN INDIVIDUAL RESIDENT VENDOR AND HAS RESIDED CONTINUOUSLY IN WEST VIRGINIA FOR FOUR (4) YEARS IMMEDIATELY PRECEDING THE DATE OF THIS CERTIFICATION; OR

( ) BIDDER IS A PARTNERSHIP, ASSOCIATION OR CORPORATION RESIDENT VENDOR AND HAS MAINTAINED ITS HEADQUARTERS OR PRINCIPAL PLACE OF BUSINESS CONTINUOUSLY IN WEST VIRGINIA FOR FOUR (4) YEARS IMMEDIATELY PRECEDING THE DATE OF THIS CERTIFICATION; OR 80% OF THE OWNERSHIP INTEREST OF BIDDER IS HELD BY ANOTHER INDIVIDUAL, PARTNERSHIP, ASSOCIATION OR CORPORATION RESIDENT VENDOR WHO HAS MAINTAINED ITS HEADQUARTERS OR PRINCIPAL PLACE

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Purchasing Division
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OF BUSINESS CONTINUOUSLY IN WEST VIRGINIA FOR FOUR (4) YEARS IMMEDIATELY PRECEDING THE DATE OF THIS CERTIFICATION; OR

( ) BIDDER IS A CORPORATION NONRESIDENT VENDOR WHICH HAS AN AFFILIATE OR SUBSIDIARY WHICH EMPLOYS A MINIMUM OF ONE HUNDRED STATE RESIDENTS AND WHICH HAS MAINTAINED ITS HEADQUARTERS OR PRINCIPAL PLACE OF BUSINESS WITHIN WEST VIRGINIA CONTINUOUSLY FOR THE FOUR (4) YEARS IMMEDIATELY PRECEDING THE DATE OF THIS CERTIFICATION.

B. APPLICATION IS MADE FOR 2.5% PREFERENCE FOR THE REASON CHECKED:

( ) BIDDER IS A RESIDENT VENDOR WHO CERTIFIES THAT, DURING THE LIFE OF THE CONTRACT, ON AVERAGE AT LEAST 75% OF THE EMPLOYEES WORKING ON THE PROJECT BEING BID ARE RESIDENTS OF WEST VIRGINIA WHO HAVE RESIDED IN THE STATE CONTINUOUSLY FOR THE TWO YEARS IMMEDIATELY PRECEDING SUBMISSION OF THIS BID;
OR
( ) BIDDER IS A NONRESIDENT VENDOR EMPLOYING A MINIMUM OF ONE HUNDRED STATE RESIDENTS OR IS A NONRESIDENT VENDOR WITH AN AFFILIATE OR SUBSIDIARY WHICH MAINTAINS ITS HEADQUARTERS OR PRINCIPAL PLACE OF BUSINESS WITHIN WEST VIRGINIA EMPLOYING A MINIMUM OF ONE HUNDRED STATE RESIDENTS WHO CERTIFIES THAT, DURING THE LIFE OF THE CONTRACT, ON AVERAGE AT LEAST 75% OF THE EMPLOYEES OR BIDDERS' AFFILIATE'S OR SUBSIDIARY'S EMPLOYEES ARE RESIDENTS OF WEST VIRGINIA WHO HAVE RESIDED IN THE STATE CONTINUOUSLY FOR THE TWO YEARS IMMEDIATELY PRECEDING SUBMISSION OF THIS BID.

BIDDER UNDERSTANDS IF THE SECRETARY OF TAX & REVENUE DETERMINES THAT A BIDDER RECEIVING PREFERENCE HAS

SEE REVERSE SIDE FOR TERMS AND CONDITIONS

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FAILED TO CONTINUE TO MEET THE REQUIREMENTS FOR SUCH PREFERENCE, THE SECRETARY MAY ORDER THE DIRECTOR OF PURCHASING TO: (A) RESCIND THE CONTRACT OR PURCHASE ORDER ISSUED; OR (B) ASSESS A PENALTY AGAINST SUCH BIDDER IN AN AMOUNT NOT TO EXCEED 5% OF THE BID AMOUNT AND THAT SUCH PENALTY WILL BE PAID TO THE CONTRACTING AGENCY OR DEDUCTED FROM ANY UNPAID BALANCE ON THE CONTRACT OR PURCHASE ORDER.

BY SUBMISSION OF THIS CERTIFICATE, BIDDER AGREES TO DISCLOSE ANY REASONABLY REQUESTED INFORMATION TO THE PURCHASING DIVISION AND AUTHORIZES THE DEPARTMENT OF TAX AND REVENUE TO DISCLOSE TO THE DIRECTOR OF PURCHASING APPROPRIATE INFORMATION VERIFYING THAT BIDDER HAS PAID THE REQUIRED BUSINESS TAXES, PROVIDED THAT SUCH INFORMATION DOES NOT CONTAIN THE AMOUNTS OF TAXES PAID NOR ANY OTHER INFORMATION DEEMED BY THE TAX COMMISSIONER TO BE CONFIDENTIAL.

UNDER PENALTY OF LAW FOR FALSE SWEARING (WEST VIRGINIA CODE 61-5-3), BIDDER HEREBY CERTIFIES THAT THIS CERTIFICATE IS TRUE AND ACCURATE IN ALL RESPECTS; AND THAT IF A CONTRACT IS ISSUED TO BIDDER AND IF ANYTHING CONTAINED WITHIN THIS CERTIFICATE CHANGES DURING THE TERM OF THE CONTRACT, BIDDER WILL NOTIFY THE PURCHASING DIVISION IN WRITING IMMEDIATELY.

BIDDER: PHYCOTECH INC

DATE: 8/25/08

SIGNED:

SEE REVERSE SIDE FOR TERMS AND CONDITIONS

SIGNATURE	TELEPHONE	DATE

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TITLE: President

* CHECK ANY COMBINATION OF PREFERENCE CONSIDERATION(S) IN EITHER "A" OR "B", OR BOTH "A" AND "B" WHICH YOU ARE ENTITLED TO RECEIVE. YOU MAY REQUEST UP TO THE MAXIMUM 5% PREFERENCE FOR BOTH "A" AND "B".
(REV. 12/00)

NOTICE

A SIGNED BID MUST BE SUBMITTED TO:

DEPARTMENT OF ADMINISTRATION
PURCHASING DIVISION
BUILDING 15
2019 WASHINGTON STREET, EAST
CHARLESTON, WV 25305-0130

THE BID SHOULD CONTAIN THIS INFORMATION ON THE FACE OF THE ENVELOPE OR THE BID MAY NOT BE CONSIDERED:

SEALED BID

BUYER: CB-23

RFQ. NO.: DEP13876

BID OPENING DATE: 08/28/2008

BID OPENING TIME: 1:30 PM

SEE REVERSE SIDE FOR TERMS AND CONDITIONS

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PLEASE PROVIDE A FAX NUMBER IN CASE IT IS NECESSARY TO CONTACT YOU REGARDING YOUR BID:

866 728 5579 or 269 983 3653

CONTACT PERSON (PLEASE PRINT CLEARLY):

ANN ST. AMAND

****** THIS IS THE END OF RFQ DEP13876 ****** TOTAL:

SEE REVERSE SIDE FOR TERMS AND CONDITIONS

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CONTRACT SPECIFICATIONS FOR PERIPHYTON SAMPLE PROCESSING AND IDENTIFICATION

AREA OF WORK / BID AWARD

The West Virginia Department of Environmental Protection, Division of Water and Waste Management (DWWM) is seeking bids for the processing and identification of diatoms and soft algae from periphyton samples collected from streams of West Virginia. Personnel from DWWM's Watershed Assessment Section will collect and preserve the samples. There are typically between 250 and 350 samples collected each year that would need processed and identified. As of 6/2008, we have a backlog of around 425 samples, of which, approximately 300 would be shipped upon finalization of contract.

Bids should be submitted by vendors in connection with the costs associated with processing (including cleaning and preparation of slides for diatoms) and identification of diatoms and soft algae from the periphyton samples.

QUALIFICATIONS

The Department of Environmental Protection's (DEP) Division of Water and Waste Management (DWWM) conducts inspections of permitted and non-permitted facilities, investigates complaints, monitors ambient quality of surface water, groundwater and sediments, performs studies, and provides water quality information to the citizens of West Virginia and other government agencies. Legal action based upon identification results is possible. Therefore, the vendor(s) selected must have a quality control program in place and meet the following qualifications:

1. Degreed biologist on staff **who performs the actual identifications**. Must have at least 2 years experience in the identification of algae samples. (Identification of organisms by non-professional personnel is strictly forbidden)

2. Capable of attending and providing expert testimony in legal proceedings, upon request.

3. Experience demonstrating ability to process and identify up to30 samples per month.

In order to verify the vendor meets the above criteria, the vendor must submit a description of how the project will be managed by the contractor, a summary of experience with sorting and identification of periphyton, a description of how the samples will be processed and identified, and a description of vendor's internal QA/QC procedures.

SCOPE

In administering and enforcing most of the pollution control laws of the state, the importance of quality control cannot be overstated. Quality control measures must be strictly adhered to in all

phases of sample collection, preservation, transportation, and analysis. The quality control and analytical processes, as they relate to the contractor's responsibility, are divided into four (4) major steps:

STEP 1 - Collection of sample from specified office.
STEP 2 - Conduct specified analysis on samples in a timely and professional manner.
STEP 3 - Establishment of continuing program to ensure the reliability of data (Quality Assurance/Quality Control).
STEP 4 - Legal Testimony

Step 1 - Collection of Samples from Specified Office

Collection of periphyton samples shall be conducted by DWWM personnel. Each sample will be a 100 ml graduated container (or similar) with periphyton scraped from 5 rocks mixed with rinse water and preserved with formalin. These will generally be total samples. There will be some split for QA purposes. The vendor will be notified of sample shipment. Costs of sample shipment to the vendor will be borne by the DWWM. Costs to return identified slides and results to the DWWM will be the sole responsibility of the successful bidder(s). The vendor shall be responsible for preservation of the sample and the internal chain of custody from the time the vendor obtains the sample until the time the analysis is accepted by the Division. The vendor shall also maintain records of the results of identification for a minimum of three (3) years.

Step 2 - Conduct Specified Analysis on Samples

Processing and Identification of Periphyton Samples shall be carried out according to the vendor's procedures as defined in response to this request.

Results of identifications shall be submitted to DWWM at a rate of at least 30 samples per month, starting 30 days from the receipt of samples or at an alternate rate that is determined acceptable by DWWM.

Analysis of samples is not deemed completed until the data has been submitted to and accepted by the DWWM. Should the DWWM not provide notice of acceptance within four weeks of the date results were mailed by the vendor, the firm may consider the data to be acceptable by the Division.

Step 3 - Quality Control

Quality control procedures should be well defined and strictly adhered to in all aspects of processing, storage, and identification. Quality control procedures must be submitted as part of this bidding process. Any cost for internal QA/QC procedures should be incorporated into the cost / sample bid.

PRIME VENDOR RESPONSIBILITIES

A vendor, who is awarded a contract, when performing work under the terms and conditions of this contract, is solely responsible for the satisfactory completion of the work. The prime vendor shall be responsible for ensuring that any subcontractors have all the necessary permits, certifications, experience and insurance to perform the work. DWWM will consider the prime vendor to be the sole point of contact with regard to authorized work under the contract; however this provision does not prohibit the DWWM from directly contacting subcontractors.

CONFIDENTIALITY

The vendor agrees that any and all data, analyses, materials, reports or other information, oral or written, prepared by the vendor with respect to this requisition shall, except for information which has been publicly available, be treated as confidential and shall not be utilized, released, published, or disclosed, by the vendor at any time for any purpose whatsoever other than to provide consultation or other service to the DWWM.

DEP reserves the right to award the contract to the two (2) lowest vendors. The second vendor would receive approximately 10% of the samples and act as a QA/QC.

SPECIFICATIONS FOR PERIPHYTON PROCESSING AND IDENTIFICATION

"Soft" (Non-Diatom) Algae – Relative Abundance and Taxa Richness

Homogenize the sample with a blender. Pipette a subsample into a Palmer counting cell. Permanent mounting techniques can be utilized if preferred. Dilute samples if cells overlap too much for counting. Identify and count 300 algal non-diatom units to the lowest taxonomic level (which should be genus and perhaps species level) at a magnification of at least 400X (higher levels of magnification are permissible). Cell units, of 10 um length, should be counted instead of individual cells for filamentous species (or measure average cells per filament based on average cell length per filament). Individual cells of colonial species should be counted when appropriate. Count live and dead (those with no cell content) diatoms separately, recording only the number of each observed in order to determine live: dead diatom ratio (identification will be done on the cleaned samples). Record numbers of non-diatom algal units on the non-diatom bench sheet that should be similar to the example provided in Appendix A of EPA's *Rapid Bioassessment Protocols for Use in Wadeable Streams and Rivers 1999* (Barbour et. al.) http://www.epa.gov/owow/monitoring/rbp/app_a.html . All undigested samples shall be archived and returned to the WVDEP at the cessation of the contract or upon request.

Calculate Biovolume / Biomass

Biomass should be calculated for the most abundant taxa (> 10% of sample) at minimum, it is fine to include measurements for all taxa. A minimum of 15 measurements (length and width) should be taken for these species. The average measurements are used to calculate the biovolume, which is then converted to biomass, assuming a specific gravity of 1. The biomass of each species is calculated based on the abundance of that species and adjusted for original sample volume.

Diatoms

Clear diatom frustules of organic and intercellular material using either 'Nitric Acid Oxidation' or 'Hydrogen Peroxide / Potassium Dichromate Oxidation'. Prepare slides and identify diatoms at 1000X to the lowest possible taxonomic level, preferably to the species or variety level, using current taxonomic references. Record all taxa encountered on the diatom bench sheet creating a species list prior to enumeration. Scan the slide until several minutes pass without producing any new taxa. For QA/QC reasons, the Contractor shall mark the beginning and end of each transect on each diatom slide using a diamond scribe, and note the length. All slides, split samples and the

digested slurry shall be archived and returned to the WVDEP at the cessation of the contract or upon request. For quantitative data, count a minimum of 600 valves recording taxa and number counted on the diatom bench sheet.

Taxonomic Resolution

Using modern literature, the contractor shall identify soft algae to genus and, if possible species, and the diatoms to the lowest possible level, which should be species and perhaps variety/subspecies. Diatoms shall be counted if they have intact frustules in the field of view. We seek a standardized level of taxonomy across all samples; at the same time we recognize that for some taxa, this goal is not reasonably achievable. These circumstances shall be noted in a comments portion of the data files.

QA/QC analysis

The Contractor shall provide the prepared diatom slides to the WVDEP in groups of approximately 30 samples, which shall include samples that are completely identified, enumerated, error-checked, data entered and verified, and shall be provided to the WVDEP with database output in electronic format. WVDEP will provide a database for this purpose. WVDEP will conduct a QA check on at least three of the first 30 samples. Sixty percent (60%) community similarity (PCS) is the lower limit in order for the Contractor to be paid for all services related to that batch of 30 samples. The Contractor shall continue to process and identify samples while the WVDEP is conducting the QA/QC analyses, unless informed otherwise. Should PCS during QA/QC be found to be between 60% - 100%, the Contractor's analysis results will be accepted and the contractor shall continue to process and identify samples in groups of 30, returning identified organisms and data sheets for each such group upon completion, until all samples have been processed. If at any point WVDEP determines that PCS is below 60%, the WVDEP will inform the Contractor in writing that the batch is not acceptable. The batch will be shipped back to the Contractor at the Contractor's expense for a second opportunity to meet the 60% PCS level within 15 days of receiving the batch. The Contractor shall expeditiously ship all counted algal samples and data sheets (bench sheets in electronic or paper format) from the returned batch to the WVDEP. If the Contractor's PCS level is greater than or equal to 60%, the Contractor shall be informed that the batch is accepted and may continue to process and identify samples with 10% of all samples undergoing QA/QC as previously noted. If the Contractor's PCS remains under 60%, or is again found to be below 60% at any time during the remainder of the contract period,

the WVDEP reserves the right to either ask the Contractor to proceed with trying to reach the 60% PCS level or terminate the contract for default.

Reporting

All taxa, counts, and biomass information should be entered into the database that WVDEP will provide. In addition, the lab will provide copies of all bench sheets generated (either paper or electronic) which includes all site information, including stream name, stream code, stream mile (if provided), date collected, collector's name, taxonomist's name, and date identified. If vendor typically calculates metrics, provide bids both with and without these calculations.

PERIPHYTON SAMPLE PROCESSING & IDENTIFICATION

Requisition # DEP13876

Company Name: PHYCOTECH INC.

Address: 620 BROAD STREET SUITE 100

City, State, Zip: ST JOSEPH MI 49085

Phone & Fax No.: 269-983-3654 866-728-5579

ITEM NO.	QUANTITY	DESCRIPTION	UNIT PRICE	AMOUNT
1.0	*700	Processing & Identification of Diatoms & Soft Algae from Periphyton Samples	$ 650.00 per sample	$ 455000.00
		SAMPLES CAN BE PROCESSED AT A RATE OF 70 SAMPLES PER YEAR, FOR QA/QC ONLY		
		TOTAL		$ 455000.00

*Quantities listed above are for bid evaluation purposes only.

RFQ No. DEP13876

STATE OF WEST VIRGINIA
Purchasing Division

PURCHASING AFFIDAVIT

VENDOR OWING A DEBT TO THE STATE:
West Virginia Code §5A-3-10a provides that: No contract or renewal of any contract may be awarded by the state or any of its political subdivisions to any vendor or prospective vendor when the vendor or prospective vendor or a related party to the vendor or prospective vendor is a debtor and the debt owed is an amount greater than one thousand dollars in the aggregate.

PUBLIC IMPROVEMENT CONTRACTS & DRUG-FREE WORKPLACE ACT:
West Virginia Code §21-1D-5 provides that: Any solicitation for a public improvement construction contract shall require each vendor that submits a bid for the work to submit at the same time an affidavit that the vendor has a written plan for a drug-free workplace policy in compliance with Article 1D, Chapter 21 of the West Virginia Code. A public improvement construction contract may not be awarded to a vendor who does not have a written plan for a drug-free workplace policy in compliance with Article 1D, Chapter 21 of the West Virginia Code and who has not submitted that plan to the appropriate contracting authority in timely fashion. For a vendor who is a subcontractor, compliance with Section 5, Article 1D, Chapter 21 of the West Virginia Code may take place before their work on the public improvement is begun.

ANTITRUST:
In submitting a bid to any agency for the state of West Virginia, the bidder offers and agrees that if the bid is accepted the bidder will convey, sell, assign or transfer to the state of West Virginia all rights, title and interest in and to all causes of action it may now or hereafter acquire under the antitrust laws of the United States and the state of West Virginia for price fixing and/or unreasonable restraints of trade relating to the particular commodities or services purchased or acquired by the state of West Virginia. Such assignment shall be made and become effective at the time the purchasing agency tenders the initial payment to the bidder.

I certify that this bid is made without prior understanding, agreement, or connection with any corporation, firm, limited liability company, partnership or person or entity submitting a bid for the same materials, supplies, equipment or services and is in all respects fair and without collusion or fraud. I further certify that I am authorized to sign the certification on behalf of the bidder or this bid.

LICENSING:
Vendors must be licensed and in good standing in accordance with any and all state and local laws and requirements by any state or local agency of West Virginia, including, but not limited to, the West Virginia Secretary of State's Office, the West Virginia Tax Department, West Virginia Insurance Commission, or any other state agencies or political subdivision. Furthermore, the vendor must provide all necessary releases to obtain information to enable the Director or spending unit to verify that the vendor is licensed and in good standing with the above entities.

CONFIDENTIALITY:
The vendor agrees that he or she will not disclose to anyone, directly or indirectly, any such personally identifiable information or other confidential information gained from the agency, unless the individual who is the subject of the information consents to the disclosure in writing or the disclosure is made pursuant to the agency's policies, procedures and rules. Vendors should visit **www.state.wv.us/admin/purchase/privacy** for the Notice of Agency Confidentiality Policies.

Under penalty of law for false swearing (West Virginia Code §61-5-3), it is hereby certified that the vendor acknowledges the information in this said affidavit and is in compliance with the requirements as stated.

Vendor's Name: PHYCOTECH INC

Authorized Signature: _____ Date: 8/25/08

Ann L. St. Amand, Ph.D., CLP

PhycoTech, Inc
620 Broad St., Ste. 100
St. Joseph, MI 49085
Voice 269.983.3654 E-Mail astamand@phycotech.com

Phycologist & Certified Lake Professional

Professional Experience

President. PhycoTech, Inc. 1990-Present. St. Joseph, Michigan. Provide identification and enumeration of suspended and attached algal, zooplankton and bacterial samples utilizing a unique, permanent mounting technique. Also provide photographic, statistical and interpretive services involving algal samples and ecological data.

Research Associate. University of Notre Dame. 1991-1995. Department of Civil Engineering/Geological Sciences. Involved in project relating composition and biomass of periphytic biolayer in artificial stream ecosystems to PCB transfer within stream sediments.

Research Associate. University of Notre Dame. 1989-1991. Department of Civil Engineering. Involved in project relating groundwater quality to surface water quality including preliminary data acquisition and grant submission. Also involved in data analysis for a collaborative project on the environmental effects of oil-field brine application for road maintenance.

Faculty. Practicum in Aquatic Ecology, University of Notre Dame Environmental Research Center. June 1990. Taught limnology section of summer field course.

Research Assistant. University of Notre Dame. 1988-1989. Identified and enumerated phytoplankton samples from three northern Wisconsin lakes.

Teaching Assistant. University of Notre Dame. 1985-1988.

Introductory Biology (majors and non-majors) - 1 sem. ea.

Invertebrate Biology - 1 sem.

Biostatistics - 2 sem.

Biological Research Applications of Computers - 1 sem.

Aquatic Ecology- Summer field course conducted at the field station - 2 summers

Field Intern. The Nature Conservancy (Indiana Chapter). November 1984-January 1985. Habitat management and landowner responsibilities within wetland and prairie habitats.

Education

Ph.D. University of Notre Dame, Notre Dame, Indiana. Aquatic Biology Program. Defense: April 12,1990. Dissertation: Mechanisms Controlling Metalimnetic Communities and the Importance of Metalimnetic Phytoplankton to Whole Lake Primary Productivity.

B.S. Purdue University, West Lafayette, Indiana. 1984. Ecology, Evolutionary, and Population Biology.

Indiana University-Purdue University at Indianapolis, Indianapolis, Indiana. 1980-1982.

Certifications

Certified Lake Professional 2003/2006
North American Lake Management Society

Honors

Secchi Disk Award. North American Lake Management Society. 2006.

Merlin Hanson Challenge Award, Cornerstone Alliance. February 2000.

Corporate Award, North American Lake Management Society. November 1999.

Best Student Paper. North American Lake Management Society. November 1988.

Graduate Student Travel Award. North American Lake Management Society. 1988.

Arthur J. Schmitt Dissertation-Year Fellowship - 1988/1989. University of Notre Dame.

Best Student Paper. North American Lake Management Society. November 1987.

IBM Travel Fund for Women. University of Notre Dame. 1987.

Grants

Heart of Cook. Education Grant. UpStream Project, Upton Middle School. May, 2007.

Florida Department of Environmental Protection: Research Grant. Joint project between PhycoTech and CyanoLab. Investigations in the biology and the ecological impact of Cylindrospermopisis raciborskii in Florida Lakes. September 2005.

National Science Foundation: Small Business Innovation Research Grant. 1997. Computerized Algal Identification System. Phase I.

National Science Foundation: Research Planning Grant. 1991. Role of the periphytic biolayer in mediating the transfer and transformation of PCB's within stream sediments.

National Science Foundation: Doctoral Dissertation Improvement Grant. 1988.

Alternate states of metalimnetic systems resulting from cascading trophic interactions.

Sigma Xi. 1988. Variation in primary productivity and biomass accumulation in metalimnetic and epilimnetic algal populations.

Indiana Academy of Science. 1987. Species-specific growth and loss rates within metalimnetic algal populations.

Presentations

St. Amand, A. and K. Wagner (Co-Chairs). 1991-Present. Algal Identification Workshop. Annual Meeting of the North American Lake Management Society.

St. Amand, A. 2007. Multiple Data Analysis Techniques and Challenges for Analyzing Phytoplankton Data from the National Lakes Survey, Summer 2007. 20th Annual National Conference Enhancing the States' Lake Management Programs: Interpreting Lake Quality Data for Diverse Audiences. April 24-27, 2007 Chicago, IL.

St. Amand, A. 2006. Occurrence and Toxicity of *Cylindrospermopsis* and other toxigenic blue-greens in the Midwest. Illinois Lake Management Association Annual Meeting. March, 2006.

St. Amand, A., Graham, J. and Jones, J. 2005. Co-Chairs. Special Session: Toxic Freshwater Cyanobacteria – Global Perspectives on North American Occurrence and Regulation. International Symposim of the North American Lake Management Society. November 9, 2005.

St. Amand, A., Dyble, J., Chapman, A., and Eilers, J. 2005. Efficacy of Molecular DNA Methods for Confirming Species Identifications on

Morphologically Variable Populations of Toxin Producing Anabaena (Nostocales). Special Session:Toxic Freshwater Cyanobacteria – Global Perspectives on North American Occurrence and Regulation. Ann St. Amand, Co-Chair. International Symposim of the North American Lake Management Society. November 9, 2005.

Eilers, J.M. and St. Amand, A. 2005. Multiple Scenarios for Fisheries to Increase Potentially Toxin Producing Cyanobacteria Populations in Selected Oregon Lakes. EPA ISOC-HAB meetings, Raleigh, NC. September 7, 2005.

St. Amand, A., Hoover, R., Jermanb, G.,and Rozanov, A. 2005. Morphology and Elemental Composition of Recent and Fossil Cyanobacteria. Proceedings of the Annual Symposium of the International Society for Optical Engineering, SPIE, July 31, 2005.

St. Amand, A. and K. Wagner. 2003 Water quality factors driving blue-green algal blooms in lakes with relatively low nutrient concentrations. Annual Meeting of the North American Lake Management Society.

St. Amand, A. 2003. Cylindrospermopsis raciborskii and its distribution changes across the US. Annual Meeting of the North American Lake Management Society.

St. Amand, A. and K. Wagner. 2003. Ecology and Control of Nuisance Algae workshop. Annual Meeting of the North American Lake Management Society.

St. Amand, A. and J. M. Eilers. 2002. Blue-green akinetes from sediment cores as a tool for assessing water quality changes. Annual Meeting of the North American Lake Management Society.

St. Amand, A. 2002. Cylindrospermopsis raciborskii: Distribution changes over the last decade across the United States and Implications for Water Quality in the Midwest. Meetings of the Midwest Aquatic Plant Management Society.

St. Amand, A. 2001. Cylindrospermopsis raciborski: Distribution, Ecology and Implications for Drinking Water Supplies and Recreational Use. Indiana Department of Environmental Management, meeting of the Task Force on Toxic Blue-Green Algae.

Wang, H., St. Amand, A., and Gray, K.A. 1992. Role of the periphytic biolayer in mediating the transfer and transformation of PCB's within stream sediments. Hazardous Waste Conference, Center for

Bioengineering and Pollution Control, University of Notre Dame.

St. Amand, A.L., P.A. Soranno, and S.R. Carpenter. 1989. Nutrient cycling dynamics associated with manipulations in fish communities. Annual Meeting of the International Association for Great Lakes Research.

St. Amand, A.L., and S.R. Carpenter. 1989. Species-specific responses to nutrient regeneration among metalimnetic and epilimnetic algae. Annual Meeting of the Ecological Society of America.

St. Amand, A.L., P.A. Soranno, and S.R. Carpenter. 1988. Nutrient deficiency indicators: Growth bioassays vs. physiological indicators for assessing ecosystem stress. Annual Meeting of the North American Lake Management Society.

St. Amand, A.L. and S.R. Carpenter. 1988. Metalimnetic and epilimnetic algae: Differences in nutrient limitation and grazing response when exposed to varying assemblages of zooplankton. 1988 Annual Meeting of the Ecological Society of America.

St. Amand, A.L. and S.R. Carpenter. 1987. Variable responses of Anabaena circinalis to grazing. Annual Meeting of the North American Lake Management Society.

St. Amand, A. and S.R. Carpenter. 1987. Metalimnetic algae: Species specific growth and loss rates. Annual Meeting of the American Society of Limnology and Oceanography.

Publications

Eilers, J. M.; Loomis, D.; Amand, A. St.: Vogel, A.; Jackson, L.; Kann, J.: Eilers, B.; Truemper, H.; Cornett, J.; Sweets, R. 2007. Biological effects of repeated fish introductions in a formerly fishless lake: Diamond Lake, Oregon, USA. Fundamental and Applied Limnology/Archiv fur Hydrobiologie, Volume 169, Number 4, pp. 265-277(13).

St. Amand, A., J. Dyble, M. Aubel, A. Chapman and J. Eilers. 2007. Efficacy of molecular DNA methods for confirming species identifications on morphologically variable populations of toxin-producing Anabaena (Nostocales). Lake and Reservoir Management. 23(2):193 - 202

St. Amand, A. and A. Chapman. 2007. Using Plankton Data. LakeLine. 27 (1):34 - 40.

Holland, T. A. St. Amand, and G. Good. 2006. Otter Lake '05—A Successful Response. LakeLine. 26(2):52 – 56.

St. Amand, A., Hoover, R., Jermanb, G.,and Rozanov, A. 2005. Morphology and Elemental Composition of Recent and Fossil Cyanobacteria. Proceedings of the Annual Symposium of the International Society for Optical Engineering, SPIE, July 31, 2005. In Press.

St. Amand, A. & Wagner, K. (2004). Stalking slime: The value of monitoring your lake. *LakeLine, 24* (1), 14-16.

Eilers, J. M., Kann, J., Cornett, J., Moser, K., & St. Amand, A. (2004). Paleolimnological evidence of change in a shallow, hypereutrophic lake: Upper Klamath Lake, Oregon USA. *Hydrobiologia, 520*, 7-18.

Eilers, J. M., & St. Amand, A. (2004). *Recent paleolimnology of Blue Lake, OR.* Report to the Oregon Department of Fish & Wildlife. Roseburg, Oregon.

Kostel J. A., Gray K. A., & St. Amand A. L. (2003). The impact of metal and organic contaminants on the structure of periphyton in lotic sediments: Observations at various scales. *International Journal of Sediment Research, 19* (2), 227-235.

St. Amand, A. (2002). Cylindrospermopsis: An invasive toxic alga. *LakeLine, 22* (1), 36-38.

Havens, K. E., Beaver, J. R., East, T. E., Rodusky, A. J., Sharfstein, B., St. Amand, A., & Steinman, A. D. (2001). Nutrient effects on producers and consumers in the littoral plankton and periphyton of a subtropical lake. *Archiv fur Hydrobiologie, 152,* 177-201.

Eilers, J. M., Beaver, J., St. Amand, A., & Cornett, J. (2001*). Historical changes of zooplankton and cyanobacteria in Diamond Lake, Oregon, based on analysis of the sediment record.* Report to the Oregon Department of Fish & Wildlife. Roseburg, Oregon.

Frost, T. M., Descy, J. P., DeStasio, B. T., Gerrish, G., Hood, J., Hurley, J. P., & St. Amand, A. L. (2000). Evaluations of phytoplankton communities using varied techniques: A multi-media comparison of lakes in Northern Wisconsin USA. *Verhandlungen Internationale Vereinigung für theoretische und angewandte Limnologie, 27,* 1023-1030.

Wang, H., Kostel, J. A., St. Amand, A. L., & Gray, K. A. (1999). The

response of a laboratory stream system to PCB exposure: Study of periphytic and sediment accumulation patterns. *Water Research, 33* (18), 3749-3761.

Kostel, J., Wang, H., St. Amand, A. L., & Gray, K. A. (1999). Use of a novel laboratory stream system to study the ecological impact of PCB's in a periphytic biolayer. *Water Research, 33* (18), 3765-3748.

Cottingham, K. L., Carpenter, S. R., & St. Amand, A. L. (1998). Responses of epilimnetic phytoplankton to experimental nutrient enrichment in three small seepage lakes. *Journal of Plankton Research, 20*, 1889-1914.

St. Amand, A. (1995). Algae: Nature's artwork. *LakeLine, 15* (3), 10-11.

St. Amand, A. L. (1994). A photographic key to the algae of the University of Notre Dame Environmental Research Center. University of Notre Dame. Ongoing. St. Amand, A. (1995). Algae: Nature's artwork. *LakeLine, 15* (3), 10-11.

St. Amand, A., & Carpenter, S. R. (1993). Plankton vertical structure. Cascading Trophic Interactions. In S. R. Carpenter & J. F. Kitchell, (Eds.), *The Trophic Cascade in Lakes.* Cambridge, UK: Cambridge University Press.

Carpenter, S. R., Morrice, J., Elser, J. J., St. Amand, A., & MacKay, N. A. (1993). Phytoplankton community dynamics. Cascading Trophic Interactions. In S. R. Carpenter & J. F. Kitchell, (Eds.), *The Trophic Cascade in Lakes.* Cambridge, UK: Cambridge University Press.

Carpenter, S. R., Morrice, J., Soranno, P. A., Elser, J. J., MacKay, N. A., & St. Amand, A. (1993). Dynamics of primary production. Cascading Trophic Interactions. In S. R. Carpenter & J. F. Kitchell, (Eds.), *The Trophic Cascade in Lakes.* Cambridge, UK: Cambridge University Press.

St. Amand, A. L. (1990). Mechanisms controlling metalimnetic communities and the importance of metalimnetic phytoplankton to whole lake primary productivity. Ph.D. dissertation, University of Notre Dame.

St. Amand, A. L., Soranno, S. A., Carpenter, S. R., & Elser, J. J. (1989). Algal nutrient deficiency: Growth bioassays versus physiological indicators. *Lake Reservoir Management, 5*, 27-35.

Elser, J. J., Goff, N. C., MacKay, N. A., St. Amand, A. L., Elser, M. M., &

Carpenter, S. R. (1987). Species-specific algal responses to zooplankton: Experimental and field observations in three nutrient-limited lakes. *Journal of Plankton Res*earch, *9*, 699-717.

Technical Skills

General

Experimental/Monitoring Design

Biostatistics

Taxonomic Skills

Phytoplankton (marine and freshwater)

Periphyton (marine and freshwater)

Core analysis for diatoms and akinetes

Macrophytes (familiarity)

Zooplankton

Benthic Macroinvertebrates (including chironomids)

Professional Societies/Journals

American Water Works Association

British Phycological Society

International Association of Diatom Research

International Phycological Society

International Society for Optical Engineering

Journal for Harmful Algae

North American Benthological Society

North American Lake Management Society (National & Michigan Chapter)

Phycological Society of America

Offices

NALMS (Administrative Council 1992-1994), NALMS (Chapters' Chair 1995-1998, Region 5 Director, 2001-2004, Secretary, 2005-2006), Michigan NALMS

(President 1999-2002, Secretary 1993-1996, 2002-2007), Chapter Representative to NALMS 1994 to present, Steering Committee WFB (1994-1996), MLSA (Science Advisory Committee 1999-2004), Moderator, Berrien County Science Olympiad, Water Quality Section (2001 to present), JTG Chair, Plankton Section, Standard Methods.

Quality Assurance Plan

Taxonomic Accuracy

Dr. Ann St. Amand, a senior level phycologist and taxonomic expert, will perform all phytoplankton, periphyton, and zooplankton identifications, enumerations, and biovolume/biomass measurements. Dr. St. Amand has published extensively in the area of algal ecology and has processed over thirteen-thousand algal and bacterial samples, and is qualified to analyze zooplankton and macroinvertebrates. Outside taxonomists will be utilized for taxonomic verifications when necessary.

All samples are initially test mounted for counting density before final mounting. Any major questionable IDs are noted in the database during counting, and indicated on the report as uncertain for taxonomic clarity. If enough sample is present, samples are sent out to other taxonomists for taxonomic confirmation. Distribution is checked on approximately every tenth sample, during the counting process. All biovolume calculations have been verified by comparing with current literature, and by comparing calculations using outside mathematical consultations.

Sample Custody

The chain-of-custody requirements for all laboratory operations for each sample (broadly interpreted to include procedures for the preparation of reagents or supplies which become an integral part of the sample, record keeping associated with sample acquisition, documentation of sample preservation, sample labeling, sample tracking to establish chain-of-custody, and shipping and packing) and laboratory analysis (i.e., laboratory coding, storage, check-out, and documentation of sample movement) will be fully documented in our data management software. Each sample received will be assigned an individual tracking number. The sample bottle, chain-of-custody, and sample log sheet, which accompanies each sample sent, are then used in conjunction with one another, to enter the samples individual tracking number and all available sample information, into our sample database, ASA. The database allows for quick and accurate tracking of each sample received by PhycoTech. Dated and initialed entries by appropriate personnel on all worksheets and in the log database are required for data validation. All information entered into ASA is fully QA/QC'd for content and accuracy. Sample receipt is confirmed with each customer. All slides are labeled with the Tracking ID, which appears on all reports, data files and in all databases associated with that sample bottle and associated data.

Counting

Microscope: There are two microscopes used to process algal samples: an Olympus BX60, research-grade compound microscope equipped with Nomarski optics (40x, 100x, 200x, 400x, and 1000x), Phase Optics (400x, 1000x), a 1.25-2X multiplier, epifluorescence (blue, green and UV Excitation), and a trinocular head for photography, with a Microfire digital camera attached.

For larger material PhycoTech also has a dissecting microscope. The BX60 is the primary microscope used for algal and zooplankton identification. There is also an Olympus BHT, research-grade compound microscope equipped with Nomarski optics (100x, 200x, 400x, and 1000x), Phase Optics (400x), epifluorescence (blue, green and UV Excitation), and a trinocular head for photography, with a Ricoh Camera Back attached using traditional slide and print film.

Data Entry: Samples are enumerated within ASA directly. ASA is a database driven program with an integrated virtual TallyMeter module, containing over 130 databases. Up to 400 taxa can be enumerated within any one sample, and the entire database currently contains over 33,000 taxa, including algae, zooplankton, macroinvertebrates and bacteria. All calculations are completed within ASA, including concentrations, biovolumes, biomasses and diversity indices. Data files are also generated by ASA and saved in Excel format, while reports are formatted and saved to pdf format utilizing Microsoft Access, including summary graphics on a per sample basis. PhycoTech can then format data files in any format required by the customer, either horizontally or vertically oriented. QA/QC on counting is a recount done on approximately every 10^{th} sample. ASA produces a QA/QC report comparing the original sample and the recount sample (quantitatively and qualitatively), including the distribution check. Samples pass that are within 10% of the QA/QC recount, quantitatively. Percent similarity may vary up to 20% on exceptionally diverse or sparse samples.

Phytoplankton: The magnification used will depend on the size of the dominant taxa and the size and number of particulates. The goal is to count at multiple magnifications in order to correctly enumerate and identify taxa present that may vary by several orders of magnitude in size. If the sample is dominated by cells below 10-20 µm or the cells are fragile and difficult to identify, the majority of counting will be completed at 400x-1000x. Measuring for biovolume includes measuring GALD and additional measurements including length, width and depth of different aspects of the colony or cell. ASA allows up to 28 separate measurements per taxa. Cell and colony shapes are approximated to a geometric figure and or figures and the appropriate calculations made. Currently, ASA has over 44 different shapes defined. From 10 up to a total of 30 natural units (sometimes higher on exceptionally variable taxa) are measured for each taxa depending on variability and number encountered.

1. Use ONE of the following methods depending on sample composition:

A. DOMINATED BY SOFT ALGAE: If the sample is dominated by soft algae greater than 10-20 µm in GALD, count a minimum of 300 natural units and 15 fields at 200x (when possible, maximum of 100 fields). In addition, count taxa below 10 µm or fragile, difficult to identify taxa at 400x (minimum of 100 natural units and 10 fields). Spread the number of fields counted evenly over the three slides provided for each sample (i.e. 30 total fields, 10 fields per slide). Counting is completed when the standard error of the mean of the total number of natural units per field is less than 10%. For large taxa (200+ µm): scan at least one

whole slide at 100x. This tiered counting method should yield a minimum of 400 natural units per sample (well over 400 cells per sample). Extremely sparse samples or samples with high particulates will yield less than 400 natural units.

B. DOMINATED BY SOFT ALGAE: If the sample is dominated by soft algae less than 10-20 µm in GALD or is dominated by fragile, difficult to identify taxa, count a minimum of 400 natural units and 15 fields at 400x (when possible, maximum of 100 fields). In addition, count taxa above 20-30 µm in GALD at 200x (minimum of 15 fields). Spread the number of fields counted evenly over the three slides provided for each sample (i.e. 30 total fields, 10 fields per slide). Counting is completed when the standard error of the mean of the total number of natural units per field is less than 10%. For large taxa (200+ µm): scan at least one whole slide at 100x. This tiered counting method should yield a minimum of 400 natural units per sample (well over 400 cells per sample). Extremely sparse samples or samples with high particulates will yield less than 400 natural units.

C. DOMINATED BY DIATOMS: If the sample is dominated by diatoms other than large, easily identified taxa (e.g. Asterionella), count a minimum of 15 fields at 1000x, and a minimum of 400 natural units total (when possible, maximum of 100 fields). In addition, count soft algae according to size distribution (see A or B above) for a minimum of 15 fields at either 200x or 400x. Spread the number of fields counted evenly over the three slides provided for each sample (i.e. 30 total fields, 10 fields per slide). Counting is completed when the standard error of the mean of the total number of natural units per field is less than 10%. For large taxa (200+ µm): scan at least one whole slide at 100x. This tiered counting method should yield a minimum of 400 natural units per sample (well over 400 cells per sample). Extremely sparse samples or samples with high particulates will yield less than 400 natural units.

*** NOTE: The goal, regardless of magnification, is to enumerate and identify a minimum of 400 natural units per sample exclusive of misc. microflagellates.***

Periphyton: The magnification used will depend on the dominant taxa. If the sample is dominated by diatoms, the majority of counting will be completed at 1000x. If the sample is dominated by soft algae, the majority of counting will be completed at 200-400x, whichever is appropriate considering cell size and particulates. The goal is to count at multiple magnifications in order to correctly enumerate and identify taxa present that may vary by several orders of magnitude in size.

The general counting method is as follows:

1. Use ONE of the following methods depending on sample composition:

A. DOMINATED BY SOFT ALGAE: If the sample is dominated by soft algae greater than 10-20 µm in GALD, count a minimum of 300 natural units and 15 fields at 200x (when possible, maximum of 100 fields). In addition, count taxa below 10 µm or fragile, difficult to identify taxa at 400x (minimum of 100 natural units and 10 fields). Spread the number of fields counted evenly over the three slides provided for each sample (i.e. 30 total fields, 10 fields per slide). Counting

is completed when the standard error of the mean of the total number of natural units per field is less than 10%. For large taxa (200+ µm): scan at least one whole slide at 100x. This tiered counting method should yield a minimum of 400 natural units per sample (well over 400 cells per sample). Extremely sparse samples or samples with high particulates will yield less than 400 natural units.

B. DOMINATED BY SOFT ALGAE: If the sample is dominated by soft algae less than 10-20 µm in GALD or is dominated by fragile, difficult to identify taxa, count a minimum of 400 natural units and 15 fields at 400x (when possible, maximum of 100 fields). In addition, count taxa above 10-20 µm GALD at 200x (minimum of 15 fields). Spread the number of fields counted evenly over the three slides provided for each sample (i.e. 30 total fields, 10 fields per slide). Counting is completed when the standard error of the mean of the total number of natural units per field is less than 10%. For large taxa (200+ µm): scan at least one whole slide at 100x. This tiered counting method should yield a minimum of 400 natural units per sample (well over 400 cells per sample). Extremely sparse samples or samples with high particulates will yield less than 400 natural units.

C. DOMINATED BY DIATOMS: If the sample is dominated by diatoms, count a minimum of 15 fields at 1000x, and a minimum of 400 natural units total (when possible, maximum of 100 fields). In addition, count soft algae according to size distribution (see A or B above) for a minimum of 15 fields at either 200x or 400x. Spread the number of fields counted evenly over the three slides provided for each sample (i.e. 30 total fields, 10 fields per slide). Counting is completed when the standard error of the mean of the total number of natural units per field is less than 10%. For large taxa (200+ µm): scan at least one whole slide at 100x. This tiered counting method should yield a minimum of 400 natural units per sample (well over 400 cells per sample). Extremely sparse samples or samples with high particulates will yield less than 400 natural units.

*** NOTE: The goal, regardless of magnification, is to enumerate and identify a minimum of 400 natural units per sample exclusive of misc. microflagellates.***

ACID CLEANING

Phytoplankton/Periphyton: If species identifications for diatoms are required or unknown diatom taxa are present, acid cleaned mounts in Naphrax are prepared according to the following procedure:

1) Take 5-20mL of sample and transfer to a clean, 250mL Pyrex beaker in the hood. Add room-temperature nitric acid to a total volume of 40-60mL.

2) Cover with a watch glass.

3) After at least 24 hours has elapsed, carefully siphon off acid using glass siphon. Dilute acid and discard down drain with lots of extra water (Let water run for a minimum of 30 minutes after discarding acid).

4) A. Transfer remaining sample to a centrifuge tube and bring volume up to 14mL with distilled water. Cap tube, mix well, and centrifuge at 3000 RPM for 5 minutes. Remove tube and carefully remove supernatant to the 2mL volume marker with a micropipetor. Bring volume back up to 14mL with distilled water, mix well, and repeat process. Complete a minimum of 6 centrifuge cycles. Check pH. If pH is lower than 7, repeat centrifuging process until the pH reaches 7.

5) B. On the final cycle, remove supernatant to the 1 mL volume marker and bring volume back to 5 mL. Mix well to suspend pellet and decant into the storage bottle. Rinse the centrifuge tube 2 more times with 5 mL of distilled water and decant into the storage bottle. The total volume of the cleaned sample should be 15 mL. If the sample is very sparse, lower final volume.

6) Using a pasture pipette, transfer enough sample to a cover slip (#1, 22mm square) to cover the entire area and place in a vibration-free area until dry.

7) Add 1 small drop of Naphrax to the cover slip and invert onto a slide. Compress the coverslip with a clean object and place in an oven (60oC) for 1-3 hours, or finish on a hot plate.

8) Ring cover slip with fingernail polish and store.

9) Identify taxa at 1000x under oil immersion. Reference taxa are identified using a diamond scribing objective and permanent ink labels.

Zooplankton: Zooplankton are enumerated at 100x to 200x, depending on the average size of animal present (structures can be viewed at 400x, if necessary). Counting procedure is consistent with Standard Methods, with the target being 200 animals. Studies requiring greater precision or focusing on diversity require a higher counting threshold. Generally, when the sample is sparse, the entire slide is counted. Measurements for biomass include length, width and depth. ASA calculates biomass on crustaceans using published length/weight regressions, and on rotifers using biovolume formulae where biovolume is then converted to biomass. ASA can also use constant weights. If requested, customers may provide custom biomass calculations for ASA to use as well.

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PhycoTech, Inc.

General Technical Approach

PhycoTech is the only commercial lab in North America to utilize a unique proprietary permanent mounting technique for archiving and preparing samples for enumeration. These mounts allow you to get further data at a later date, as well as maintain a permanent archive of the sample that is easily stored, maintains fluorescence, and does not degrade with time (100+ years). Permanent algal mounts allow archiving of diatoms AND soft algae. All periphyton samples to species include both HPMA mounts for the whole sample and Naphrax, acid cleaned mounts, for diatom identification to species level. Zooplankton samples are also permanently mounted using a slightly different process. Our algal taxonomist, Dr. Ann St. Amand, has over 23 years of experience and has processed over 25,000 periphyton, phytoplankton, bacteria and zooplankton samples from both freshwater and marine systems. Dr. St. Amand is the only person who enumerates algal and zooplankton samples at PhycoTech, ensuring data integrity and consistency. Our In-house key and publication library numbers in the thousands, including the most current references. We have processed several state wide surveys in the Mid-West, West and Florida for phytoplankton and periphyton, each comprised of several hundreds of samples. PhycoTech also consults with Federal and State Agencies, including the Corps of Engineers, on experimental design and QA/QC issues. We process samples for general water quality, as well as the determination of exotic, toxic or taste and odor producing blue-green and chrysophyte algae. PhycoTech has extensive experience enumerating Prymesium parvum, a difficult to identify, toxic haptophyte from the Southwestern United States.

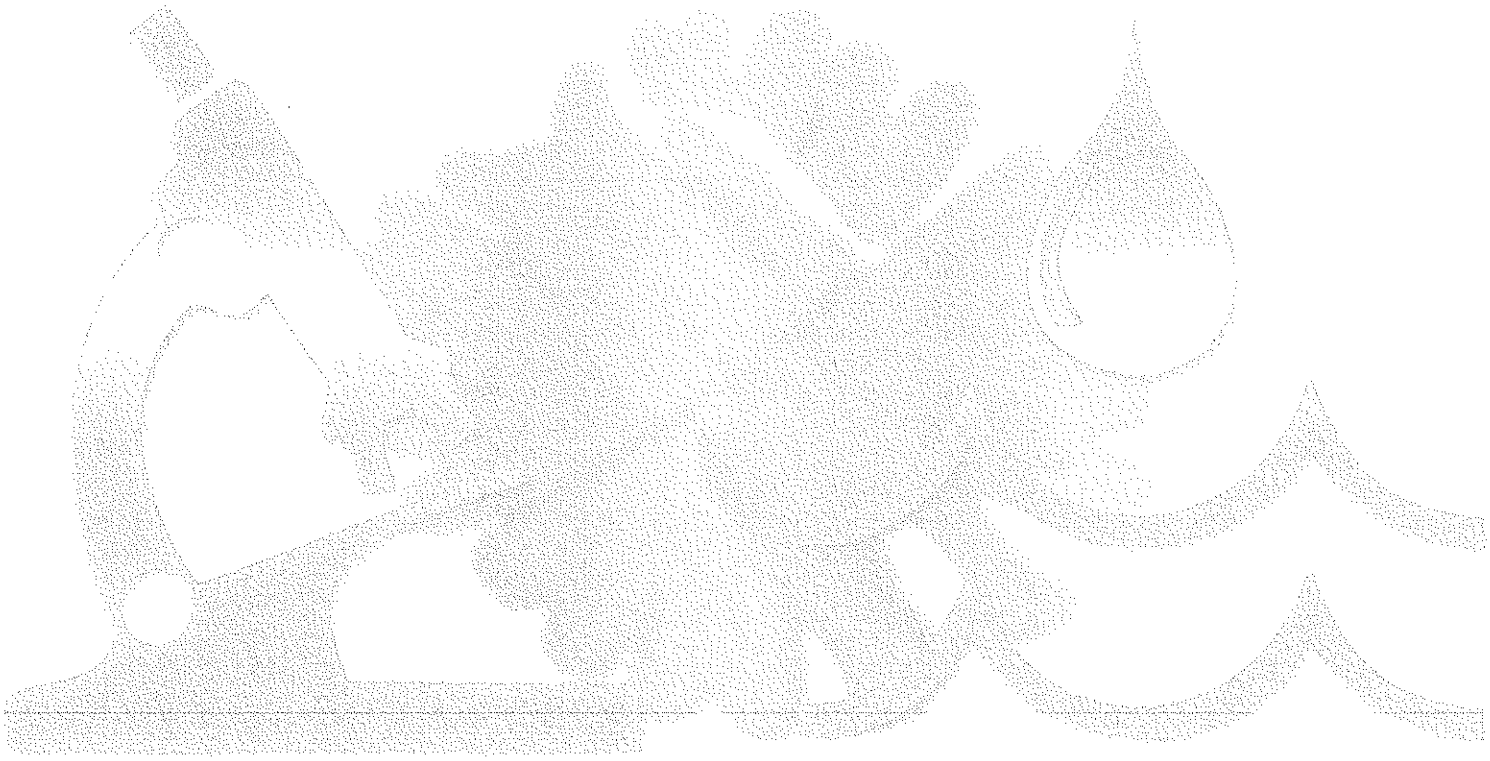
There are three state of the art microscopes used to process algal and zooplankton samples: an Olympus BX51, research-grade compound microscope equipped with Nomarski optics (40x, 100x, 200x, 400x, and 1000x), Phase Optics (200x, 400x, 1000x), Polarized light for zebra mussel velliger counts, and Reflective Incident Light, (1000x) a 1.25-2X multiplier, epifluorescence (blue, green and UV Excitation), and a trinocular head for photography, with a SpotFlex digital camera attached; an Olympus BX60, research-grade compound microscope equipped with Nomarski optics (40x, 100x, 200x, 400x, and 1000x), Phase Optics (400x, 1000x), a 1.25-2X multiplier, epifluorescence (blue,

green and UV Excitation), and a trinocular head for photography, with a Microfire digital camera attached, and an Olympus BHT, research-grade compound microscope equipped with Nomarski optics (100x, 200x, 400x, and 1000x), Phase Optics (400x), epifluorescence (blue, green and UV Excitation), and a trinocular head for photography, with a Ricoh Camera Back attached using traditional slide and print film. For larger material PhycoTech also has a dissecting microscope. We have access to Notre Dame's SEM facility as well. Our computer network is a newly installed Novel Network connected to 6 new 2.8+ Gig workstations, with special adaptations for graphics and extra memory for efficiently handling our new data management system, Aquatic Sample Analysis (ASA) System. All count related data is backed up daily to different media, with on-site and off-site copies weekly.

PhycoTech is now utilizing its proprietary data management software, ASA. This unique, powerful program not only tracks samples from receipt to data delivery within the same software program (every processing step is documented with initials and date, from Login to Analysis), but also provides significantly more information for each sample. With ASA, we are able to provide not only biovolume estimates, but volume and surface area estimates as well. Our biovolume, volume and area formulas are the most complete set available commercially, drawn from a variety of sources including current primary literature (See our Technical Approach), custom calculations designed in-house for complicated morphologies (e.g. Ceratium) and independently derived calculations from an Outside Engineering Firm that specializes in volumetric studies (e.g. area of a prolate, oblate ellipsoid). We also now provide data summaries on phyla, division, class down to taxa level automatically, depending on the analysis requested. In addition, our new program has the capability to calculate over 72 different diversity indices and summary statistics, including Shannon, Maragalef, Alpha and Berger Parker Diversity measures, Species Richness and Evenness, Pollution Tolerance for diatoms, Environmental Tolerance for algae, Siltation Index for diatoms, Pollution Tolerance for diatoms, Palmer Index, ACC:CMN for diatoms, in addition to others. All taxonomic information from organism down to coloniality and structure is provided in the data set. All indices are calculated on an abundance (both Natural units/mL and Cells/mL) and total biovolume, biomass, volume and area basis, if biovolume/biomass is measured. QA/QC reports are generated from within the program,

comparing dominant taxa, reporting distribution checks and doing similarity calculations between the original sample and QA/QC sample. All slides are labeled with a unique Tracking ID code that appears on every report, data file and database generated within the laboratory.

Reports are provided in pdf format with summary graphics by group for each sample. Data files are provided in Excel format or other spreadsheet or database formats requested by the customer.



PhycoTech, Inc.

Algae

The HPMA method for producing algal sample slides provides an optically clear background while permanently infiltrating and preserving the sample for archival purposes (See references). Mounting distortion is minimal and the method provides the advantage of being able to go 100x to 1000x on the same specimen. Wet sample is always maintained in case clarification of identification is necessary. We strongly encourage our customers to use glutaraldehyde (final concentration of 0.25-0.50 %) for preservation of algal samples. It offers minimal distortion and allows the use of epifluorescence on algal samples while counting, which can dramatically improve the final results.

1) GENERAL PROTOCOL FOR MAKING PERMANENT ALGAL MOUNTS USING HPMA

EQUIPMENT:

Bunsen burner
Beaker tongs
Ice bath
Pyrex beakers (150 ML)
2 Dropper bottles
Mixed ester nitrocellulose filters (0.45 µm, 25 mm, plain)
Glass slides (25 mm x 75 mm)
Avery Laser Labels: #2181
Glass coverslips (25 mm x 25 mm, #1 or #1.5)
Full view series support/drying racks (102 pin)
Graduate cylinders
Dumont forceps
Glass filter towers (25 mL)
Rubber stoppers (#2, #10)
Filtration Manifold (6 station)
Vacuum pump (plus appropriate plumbing, 25-50 mm Hg)

Drying oven (60"C, not forced air)

Hood

REAGENTS:

HPMA (2-hydroxypropyl methacrylate)

Catalyst (azo-bis-iso-butyronitrile)

Iodine

Glutaraldehyde (25%, general grade)

Distilled water

CAUTION: Store HPMA and catalyst in refrigerator. Keep glutaraldehyde under hood.

METHOD:

SAMPLE:

1. Add enough glutaraldehyde to bring the final concentration to approximately 0.25% to 0.5% (for periphyton samples or "bloom tows", increase the final concentration to approximately 0.5%-1%). Keep the sample dark and refrigerate if possible.
2. Remove the sample from the refrigerator and let it warm to room temperature before mounting.

RESIN:

1. Prepare ice bath in plastic tub.
2. Measure 25 mL of HPMA and 0.025 g of catalyst into a 150 mL beaker.
3. Deal with HPMA under hood and use gloves for both HPMA and catalyst.
4. Under hood, light Bunsen burner and set to high flame.

Heat HPMA (with catalyst added) until you see density currents starting to form. Cool mixture by swirling in ice bath, and return to flame. **DO NOT LET MIXTURE BOIL!!!!!.** Keep heating and cooling, alternately, until the mixture is approximately the thickness of Kayro syrup. Make sure the mixture is

cool when it reaches this point or it will polymerize further. Transfer to a clean, glass jar for storage until usage. The entire procedure takes 1 to 2 hours, depending on how brave you are. ***CAUTION!!*** **THIS REACTION IS EXOTHERMIC ONCE IT REACHES A CERTAIN TEMPERATURE AND WILL TAKE PLACE ALMOST EXPLOSIVELY IF YOU LET IT GET TOO HOT. THE FUMES ARE TOXIC. KEEP WATER OUT OF THE PRE-POLYMER.**

NOTE: Wash beakers in ethanol by letting them soak for 24 to 48 hours twice; wash with soap and rinse with distilled water. Be careful to keep dust out of the beakers when making the resin.

Fill 2 amber dropper bottles with resin. Add crystalline iodine to one of the bottles until the resin is nearly opaque. The iodine-resin will be slightly thicker than normal resin. (Resin is light sensitive -- be sure to cover the extra resin with foil.)

SLIDES:

MAKE THREE SLIDES FOR EACH SAMPLE -- SHAKE SAMPLE WELL (100 TIMES-phyt. or 200 TIMES-peri.). Use Millipore 6-place stainless steel manifold and Millipore Filtration Towers.

1. Put membrane filters onto filtration bases and wet with distilled water. Drain excess water through filter. If filter has any opaque areas (very white when wet), replace with another filter.
2. Assemble filter towers.
3. Measure out phytoplankton sample using micropipetor or macropipetor (use graduate cylinder for very dilute samples, e.g. 30+ mL). For periphyton samples, remove sample with micropipetor (usually from 0.05-0.5 mL) and dilute to 10 mL in a graduated cylinder with distilled water. Agitate to mix. Choose sample volume so that each field at 200x contains approximately 20-30 cells.
4. Add sample to the tower and open valve. For periphyton samples or large phytoplankton samples using cylinders, rinse graduate cylinder into tower. Filter sample until water just clears the filter surface. Close valve and remove filtration tower just after the water disappears from the inner edge of the tower.

5. Place filter, FACE down, on a cover slip (# 1.5). Be careful to avoid bubbles under the filter.
6. Samples:
    1. Samples preserved in glutaraldehyde:
        1. 3 slides: Add 1-2 drops of clear resin to the back of the filter, and rotate the cover slip until the resin covers the back of the filter.
    2. Samples preserved in lugols:
        1. 3 slides: Add 1-2 drops of the iodine-resin to the back of the filter, and rotate the cover slip until the resin covers the back of the filter.
7. Place cover slips on the drying rack and place in drying oven for 12 to 24 hours.
8. Remove cover slips from oven. Add 1 drop of resin to the filter side of the cover slip and attach to a labeled slide. Add as little resin as possible to cover the filter surface!!!! The less resin, the faster it will polymerize and the better the prep.
9. Put slides in the oven and let polymerize for approximately 24 hours. If the resin is not completely polymerized, replace and heat for as long as 2-3 days. Make sure that the slides are completely polymerized before you store them or they will run and/or evaporate!!!! And believe me, its a mess!!!!
10. Label slides with ASA generated labels. All slides are labeled with the Tracking ID, which appears on all reports, data files and in all databases associated with that sample bottle and associated data.

Zooplankton

The HPMA method for producing zooplankton sample slides provides an optically clear background while permanently infiltrating and preserving the sample for archival purposes (See references). Mounting distortion is minimal. Wet sample is always maintained in case clarification of identification is necessary. Lignin Pink Double Stain allows for better visualization of animals and highlights critical morphological structures necessary for identification. Preferred preservative for zooplankton is EtOH. Lugol's iodine can also be used, but sometimes interferes with staining and obscures structures.

2) GENERAL PROTOCOL FOR MAKING PERMANENT ZOOPLANKTON MOUNTS USING HPMA

EQUIPMENT:

Bunsen burner

Beaker tongs

Ice bath

Pyrex beakers (150 ML)

2 Dropper bottles

Mixed ester nitrocellulose filters (5.0 µm, 47 mm, plain)

Analyslide (47 mm)

Laser Labels: 1½ x ¾ inch

Full view series support/drying racks (102 pin)

Graduate cylinders

Dumont forceps

Lignin Pink Double Stain

Glass filter tower (250 mL)

Filter Flask

Rubber stopper (#8)

Glass Microanalysis Filter Holder 47 mm disc

Vacuum hand pump

Drying oven (43°C, not forced air)

Hood

REAGENTS:

HPMA (2-hydroxypropyl methacrylate)

Catalyst (azo-bis-iso-butyronitrile)

Iodine

Alcohol (70% ETOH)

Distilled water

CAUTION: Store HPMA and catalyst in refrigerator.

METHOD:

SAMPLE:

1. Add enough alcohol to bring the final concentration to approximately 70%, or Lugols until a dark tea color.

RESIN:

1. Prepare ice bath in plastic tub.
2. Measure 25 mL of HPMA and 0.025 g of catalyst into a 150 mL beaker.
3. Deal with HPMA under hood and use gloves for both HPMA and catalyst.
4. Under hood, light Bunsen burner and set to high flame.

Heat HPMA (with catalyst added) until you see density currents starting to form. Cool mixture by swirling in ice bath, and return to flame. **DO NOT LET MIXTURE BOIL!!!!!.** Keep heating and cooling, alternately, until the mixture is approximately the thickness of Kayro syrup. Make sure the mixture is cool when it reaches this point or it will polymerize further. Transfer to a clean, glass jar for storage until usage. The entire procedure takes 2 to 1 hour, depending on how brave you are. ***CAUTION!!*** **THIS REACTION IS EXOTHERMIC ONCE IT REACHES A CERTAIN TEMPERATURE AND WILL TAKE PLACE ALMOST EXPLOSIVELY IF YOU LET IT GET TOO HOT. THE FUMES ARE TOXIC. KEEP WATER OUT OF THE PRE-POLYMER.**

NOTE: Wash beakers in ethanol by letting them soak for 24 to 48 hours twice; wash with soap and rinse with distilled water. Be careful to keep dust out of the beakers when making the resin.

Fill 2 amber dropper bottles with resin. Add crystalline iodine to one of the bottles until the resin is nearly opaque. The iodine-resin will be slightly thicker than normal resin. (Resin is light sensitive -- be sure to cover the extra resin with foil.)

SLIDES:

MAKE ONE SLIDE FOR EACH SAMPLE -- SHAKE SAMPLE GENTLY 50 TIMES, if necessary, split sample with a Folsom plankton splitter.

1. Put membrane filter onto filtration base and wet with distilled water. Drain excess water through filter. If filter has any opaque areas (very white when wet), replace with another filter.
2. Assemble filter tower.
3. Measure out zooplankton sample using graduate cylinder. Choose sample volume so that each field at 100x contains approximately 5-10 animals.
4. Add one drop of Lignin Pink to graduated cylinder for every 5mL of sample. Let sample sit for 15 minutes.

5. Place entire contents of graduated cylinder into filter tower. Rinse graduate cylinder into tower twice. Filter sample (using vacuum hand pump) until water just clears the filter surface. Remove filtration tower just after the water disappears from the inner edge of the tower.
6. Place filter, FACE up, on analyslide (47 mm). Be careful to avoid bubbles under the filter.
7. Add 8-10 drops of clear resin to the filter, and rotate the analyslide until the resin covers the whole filter.
8. Place analyslide on the drying rack and place in drying oven for 12 to 24 hours.
9. Remove analyslide from oven. Add just enough resin to the filter to cover the filter surface!!!! The less resin, the faster it will polymerize and the better the prep.
10. Put slides in the oven and let polymerize for approximately 24 hours. If the resin is not completely polymerized, replace and heat for as long as 2-3 days. Make sure that the slides are completely polymerized before you store them or they will run and/or evaporate!!!! And believe me, it's a mess!!!!
11. Label slides with ASA generated labels. All slides are labeled with the Tracking ID, which appears on all reports, data files and in all databases associated with that sample bottle and associated data.

Quality Assurance Plan

Taxonomic Accuracy

Dr. Ann St. Amand, a senior level phycologist and taxonomic expert, will perform all phytoplankton, periphyton, and zooplankton identifications, enumerations, and biovolume/biomass measurements. Dr. St. Amand has published extensively in the area of algal ecology and has processed over twenty-five thousand algal and bacterial samples, and is qualified to analyze zooplankton and macroinvertebrates. Outside taxonomists will be utilized for taxonomic verifications when necessary.

All samples are initially test mounted for counting density before final mounting. Any major questionable IDs are noted in the database during counting, and indicated on the report as uncertain for taxonomic clarity. If enough sample is present, samples are sent out to other taxonomists for taxonomic confirmation. Distribution is checked on approximately every tenth sample, during the counting process. All biovolume calculations have been verified by comparing with current literature, and by comparing calculations using outside mathematical consultations.

Sample Custody

The chain-of-custody requirements for all laboratory operations for each sample (broadly interpreted to include procedures for the preparation of reagents or supplies which become an integral part of the sample, record keeping associated with sample acquisition, documentation of sample preservation, sample labeling, sample tracking to establish chain-of-custody, and shipping and packing) and laboratory analysis (i.e., laboratory coding, storage, check-out, and documentation of sample movement) will be fully documented in our data management software. Each sample received will be assigned an individual tracking number. The sample bottle, chain-of-custody, and sample log sheet, which accompanies each sample sent, are then used in conjunction with one another, to enter the samples individual tracking number and all available sample information, into our sample database, ASA. The database allows for quick and accurate tracking of each sample received by PhycoTech. Dated and initialed entries by appropriate personnel on all worksheets and in the log database are required for data validation. All information entered into ASA is fully QA/QC'd for content and accuracy. Sample receipt is confirmed with each customer. All slides are labeled with the Tracking ID, which appears on all reports, data files and in all databases associated with that sample bottle and associated data.

Counting

Microscope: There are two microscopes used to process algal samples: Our primary microscope, an Olympus BX51, research-grade compound microscope equipped with Brightfield optics(40x, 100x, 200x, 400x, 1000x), Nomarski optics (100x, 200x, 400x, and 1000x), Phase Optics (200x, 400x, 1000x), a 1.25-2X multiplier, epifluorescence (blue, green and UV Excitation), and a trinocular head for photography, with a SpotFlex digital camera attached. For larger material

PhycoTech also has a dissecting microscope. The BX60 is a secondary microscope with similar optics to the BX51, used for algal and zooplankton identification as a back-up microscope. There is also an Olympus BHT, research-grade compound microscope equipped with Nomarski optics (100x, 200x, 400x, and 1000x), Phase Optics (400x), epifluorescence (blue, green and UV Excitation), and a trinocular head for photography, with a Ricoh Camera Back attached using traditional slide and print film.

Data Entry: Samples are enumerated within ASA directly. ASA is a database driven program with an integrated virtual TallyMeter module, containing over 130 databases. Up to 400 taxa can be enumerated within any one sample, and the entire database currently contains over 33,000 taxa, including algae, zooplankton, macroinvertebrates and bacteria. All calculations are completed within ASA, including concentrations, biovolumes, biomasses and diversity indices. Data files are also generated by ASA and saved in Excel format, while reports are formatted and saved to pdf format utilizing Microsoft Access, including summary graphics on a per sample basis. PhycoTech can then format data files in any format required by the customer, either horizontally or vertically oriented. QA/QC on counting is a recount done on approximately every 10^{th} sample. ASA produces a QA/QC report comparing the original sample and the recount sample (quantitatively and qualitatively), including the distribution check. Samples pass that are within 10% of the QA/QC recount, quantitatively. Percent similarity may vary up to 20% on exceptionally diverse or sparse samples.

Phytoplankton: The magnification used will depend on the size of the dominant taxa and the size and number of particulates. The goal is to count at multiple magnifications in order to correctly enumerate and identify taxa present that may vary by several orders of magnitude in size. If the sample is dominated by cells below 10-20 µm or the cells are fragile and difficult to identify, the majority of counting will be completed at 400x-1000x. Measuring for biovolume includes measuring GALD and additional measurements including length, width and depth of different aspects of the colony or cell. ASA allows up to 28 separate measurements per taxa. Cell and colony shapes are approximated to a geometric figure and or figures and the appropriate calculations made. Currently, ASA has over 44 different shapes defined. From 10 up to a total of 30 natural units (sometimes

higher on exceptionally variable taxa) are measured for each taxa depending on variability and number encountered.

1. Use ONE of the following methods depending on sample composition:

A. DOMINATED BY SOFT ALGAE: If the sample is dominated by soft algae greater than 10-20 µm in GALD, count a minimum of 300 natural units and 15 fields at 200x (when possible, maximum of 100 fields). In addition, count taxa below 10 µm or fragile, difficult to identify taxa at 400x (minimum of 100 natural units and 10 fields). Spread the number of fields counted evenly over the three slides provided for each sample (i.e. 30 total fields, 10 fields per slide). Counting is completed when the standard error of the mean of the total number of natural units per field is less than 10%. For large taxa (200+ µm): scan at least one whole slide at 100x. This tiered counting method should yield a minimum of 400 natural units per sample (well over 400 cells per sample). Extremely sparse samples or samples with high particulates will yield less than 400 natural units.

B. DOMINATED BY SOFT ALGAE: If the sample is dominated by soft algae less than 10-20 µm in GALD or is dominated by fragile, difficult to identify taxa, count a minimum of 400 natural units and 15 fields at 400x (when possible, maximum of 100 fields). In addition, count taxa above 20-30 µm in GALD at 200x (minimum of 15 fields). Spread the number of fields counted evenly over the three slides provided for each sample (i.e. 30 total fields, 10 fields per slide). Counting is completed when the standard error of the mean of the total number of natural units per field is less than 10%. For large taxa (200+ µm): scan at least one whole slide at 100x. This tiered counting method should yield a minimum of 400 natural units per sample (well over 400 cells per sample). Extremely sparse samples or samples with high particulates will yield less than 400 natural units.

C. DOMINATED BY DIATOMS: If the sample is dominated by diatoms other than large, easily identified taxa (e.g. Asterionella), count a minimum of 15 fields at 1000x, and a minimum of 400 natural units total (when possible, maximum of 100 fields). In addition, count soft algae according to size distribution (see A or B above) for a minimum of 15 fields at either 200x or 400x. Spread the number of fields counted evenly over the three slides provided for each sample (i.e. 30 total fields, 10 fields per slide). Counting is completed when the standard error of the mean of the total number of

natural units per field is less than 10%. For large taxa (200+ µm): scan at least one whole slide at 100x. This tiered counting method should yield a minimum of 400 natural units per sample (well over 400 cells per sample). Extremely sparse samples or samples with high particulates will yield less than 400 natural units.

*** NOTE: The goal, regardless of magnification, is to enumerate and identify a minimum of 400 natural units per sample exclusive of misc. microflagellates.***

Periphyton: The magnification used will depend on the dominant taxa. If the sample is dominated by diatoms, the majority of counting will be completed at 1000x. If the sample is dominated by soft algae, the majority of counting will be completed at 200-400x, whichever is appropriate considering cell size and particulates. The goal is to count at multiple magnifications in order to correctly enumerate and identify taxa present that may vary by several orders of magnitude in size.

The general counting method is as follows:

1. Use ONE of the following methods depending on sample composition:

A. DOMINATED BY SOFT ALGAE: If the sample is dominated by soft algae greater than 10-20 µm in GALD, count a minimum of 300 natural units and 15 fields at 200x (when possible, maximum of 100 fields). In addition, count taxa below 10 µm or fragile, difficult to identify taxa at 400x (minimum of 100 natural units and 10 fields). Spread the number of fields counted evenly over the three slides provided for each sample (i.e. 30 total fields, 10 fields per slide). Counting is completed when the standard error of the mean of the total number of natural units per field is less than 10%. For large taxa (200+ µm): scan at least one whole slide at 100x. This tiered counting method should yield a minimum of 400 natural units per sample (well over 400 cells per sample). Extremely sparse samples or samples with high particulates will yield less than 400 natural units.

B. DOMINATED BY SOFT ALGAE: If the sample is dominated by soft algae less than 10-20 µm in GALD or is dominated by fragile, difficult to identify taxa, count a minimum of 400 natural units and 15 fields at 400x (when possible, maximum of 100 fields). In addition, count taxa above 10-20 µm GALD at 200x (minimum of 15 fields). Spread the number of fields counted evenly over the three slides provided for each sample (i.e. 30 total fields, 10 fields per slide). Counting is completed when

the standard error of the mean of the total number of natural units per field is less than 10%. For large taxa (200+ µm): scan at least one whole slide at 100x. This tiered counting method should yield a minimum of 400 natural units per sample (well over 400 cells per sample). Extremely sparse samples or samples with high particulates will yield less than 400 natural units.

C. DOMINATED BY DIATOMS: If the sample is dominated by diatoms, count a minimum of 15 fields at 1000x, and a minimum of 400 natural units total (when possible, maximum of 100 fields). In addition, count soft algae according to size distribution (see A or B above) for a minimum of 15 fields at either 200x or 400x. Spread the number of fields counted evenly over the three slides provided for each sample (i.e. 30 total fields, 10 fields per slide). Counting is completed when the standard error of the mean of the total number of natural units per field is less than 10%. For large taxa (200+ µm): scan at least one whole slide at 100x. This tiered counting method should yield a minimum of 400 natural units per sample (well over 400 cells per sample). Extremely sparse samples or samples with high particulates will yield less than 400 natural units.

*** NOTE: The goal, regardless of magnification, is to enumerate and identify a minimum of 400 natural units per sample exclusive of misc. microflagellates.***

ACID CLEANING

Phytoplankton/Periphyton: If species identifications for diatoms are required or unknown diatom taxa are present, acid cleaned mounts in Naphrax are prepared according to the following procedure:

1) Take 5-20mL of sample and transfer to a clean, 250mL Pyrex beaker in the hood. Add room-temperature nitric acid to a total volume of 40-60mL.
2) Cover with a watch glass.
3) After at least 24 hours has elapsed, carefully siphon off acid using glass siphon. Dilute acid and discard down drain with lots of extra water (Let water run for a minimum of 30 minutes after discarding acid).
4) A. Transfer remaining sample to a centrifuge tube and bring volume up to 14mL with distilled water. Cap tube, mix well, and centrifuge at 3000 RPM for 5 minutes. Remove

tube and carefully remove supernatant to the 2mL volume marker with a micropipetor. Bring volume back up to 14mL with distilled water, mix well, and repeat process. Complete a minimum of 6 centrifuge cycles. Check pH. If pH is lower than 7, repeat centrifuging process until the pH reaches 7.

4) B. On the final cycle, remove supernatant to the 1 mL volume marker and bring volume back to 5 mL. Mix well to suspend pellet and decant into the storage bottle. Rinse the centrifuge tube 2 more times with 5 mL of distilled water and decant into the storage bottle. The total volume of the cleaned sample should be 15 mL. If the sample is very sparse, lower final volume.
5) Using a pasture pipette, transfer enough sample to a cover slip (#1, 22mm square) to cover the entire area and place in a vibration-free area until dry.
6) Add 1 small drop of Naphrax to the cover slip and invert onto a slide. Compress the coverslip with a clean object and place in an oven (60oC) for 1-3 hours, or finish on a hot plate.
7) Ring cover slip with fingernail polish and store.
8) Identify taxa at 1000x under oil immersion. Reference taxa are identified using a diamond scribing objective and permanent ink labels.

Zooplankton: Zooplankton are enumerated at 100x to 200x, depending on the average size of animal present (structures can be viewed at 400x, if necessary). Counting procedure is consistent with Standard Methods, with the target being 200 animals. Studies requiring greater precision or focusing on diversity require a higher counting threshold. Generally, when the sample is sparse, the entire slide is counted. Measurements for biomass include length, width and depth. ASA calculates biomass on crustaceans using published length/weight regressions, and on rotifers using biovolume formulae where biovolume is then converted to biomass. ASA can also use constant weights. If requested, customers may provide custom biomass calculations for ASA to use as well.

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Olrik, K., et. al. 1998. Methods for Quantitative Assessment of Phytoplankton in Freshwaters, part I. Naturvårdsverket, Stockholm.

Hillebrand,H., et. al. 1999. Biovolume Calculation for Pelagic and Benthic Microalgae. Journal of Phycology. 35: 403-424.

Standard Methods for Examination of Water and Wastewater, 18th ed., 2005. American Public Health Association, 1015 18th Street, N.W., Washington, DC 20036.

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Algae Analysis with Biovolume Estimates
Report and Data Set

Customer ID: 000

Tracking Code:	050001-000	**Sample ID:**	Phytoplankton Grab with Biovolume	**Replicate:**	1
Customer ID:	000	**Sample Date:**	11/11/2004	**Sample Level:**	Composite
Job ID:	1	**Station:**	Sample Station	**Sample Depth:**	0
System Name:	Sample Lake	**Site:**	Sample Site	**Preservative:**	Lugols
Report Notes:	Sample Report Data				

Division: Bacillariophyta

Taxa ID	Genus	Species	Subspecies	Variety	Form	Morph	Structure	GALD µm	Count NU/ml	Relative Count	Total Biovolume µm^3 /ml	Relative Total Biovolume
1076	*Cyclotella*	*meneghiniana*	.	.	.	.	Vegetative	12.00	4.389	0.03	2,978.261	0.01
Summary for Division ~ Bacillariophyta (1 detail record)							**Sum Total Bacillariophyta**		4.389	0.03	2,978.261	0.01

Division: Chlorophyta

Taxa ID	Genus	Species	Subspecies	Variety	Form	Morph	Structure	GALD µm	Count NU/ml	Relative Count	Total Biovolume µm^3 /ml	Relative Total Biovolume
1000031	*Ankistrodesmus*	*falcatus*	.	.	.	straight	Vegetative	24.00	4.389	0.03	176.490	0.00
2683	**Chlorococcaceae*	*spp*	.	.	.	2-9.9 um spherical	Vegetative	12.00	8.778	0.05	7,942.030	0.02
2491	*Schroederia*	*judayi*	.	.	.	.	Vegetative	40.00	4.389	0.03	294.149	0.00
2031	*Ankistrodesmus*	*falcatus*	.	.	.	monoraphidiod	Vegetative	20.00	4.389	0.03	639.916	0.00
Summary for Division ~ Chlorophyta (4 detail records)							**Sum Total Chlorophyta**		21.945	0.13	9,052.584	0.03

Division: Chrysophyta

Taxa ID	Genus	Species	Subspecies	Variety	Form	Morph	Structure	GALD µm	Count NU/ml	Relative Count	Total Biovolume µm^3 /ml	Relative Total Biovolume
1180	*Mallomonas*	*spp*	.	.	.	.	Vegetative	24.00	21.945	0.13	17,648.955	0.05
7111	*Gonyostomum*	*semen*	.	.	.	.	Vegetative	55.00	1,645.851	9.88	34,939,415.992	97.85
1323	*Synura*	*spp*	.	.	.	.	Vegetative	8.00	18.175	0.11	867.915	0.00
Summary for Division ~ Chrysophyta (3 detail records)							**Sum Total Chrysophyta**		1,685.971	10.12	34,957,932.862	97.91

Division: Cryptophyta

Taxa ID	Genus	Species	Subspecies	Variety	Form	Morph	Structure	GALD µm	Count NU/ml	Relative Count	Total Biovolume µm^3 /ml	Relative Total Biovolume
3015	*Cryptomonas*	*erosa*	.	.	.	.	Vegetative	17.00	122.890	0.74	78,758.462	0.22
3069	*Cryptomonas*	*rostratiformis*	.	.	.	.	Vegetative	32.00	4.389	0.03	21,178.746	0.06
Summary for Division ~ Cryptophyta (2 detail records)							**Sum Total Cryptophyta**		127.279	0.76	99,937.208	0.28

Division: Cyanophyta

Taxa ID	Genus	Species	Subspecies	Variety	Form	Morph	Structure	GALD µm	Count NU/ml	Relative Count	Total Biovolume µm^3 /ml	Relative Total Biovolume
4285	*Synechocystis*	*spp*	.	.	.	>1 um spherical	Vegetative	1.00	14,722.066	88.38	7,708.474	0.02
Summary for Division ~ Cyanophyta (1 detail record)							**Sum Total Cyanophyta**		14,722.066	88.38	7,708.474	0.02

Division: Euglenophyta

Taxa ID	Genus	Species	Subspecies	Variety	Form	Morph	Structure	GALD µm	Count NU/ml	Relative Count	Total Biovolume µm^3 /ml	Relative Total Biovolume
5020	*Euglena*	*spp*	.	.	.	.	Vegetative	90.67	30.723	0.18	334,234.442	0.94
5023	*Euglena*	*acus*	.	.	.	.	Vegetative	120.00	13.167	0.08	20,958.135	0.06
118570	*Lepocinclis*	*glabra*	.	.	.	.	Vegetative	20.00	8.778	0.05	18,016.642	0.05
5030	*Phacus*	*spp*	.	.	.	.	Vegetative	37.33	13.167	0.08	78,537.851	0.22
5036	*Phacus*	*helikoides*	.	.	.	.	Vegetative	104.00	4.389	0.03	132,367.164	0.37
Summary for Division ~ Euglenophyta (5 detail records)							**Sum Total Euglenophyta**		70.223	0.42	584,114.233	1.64

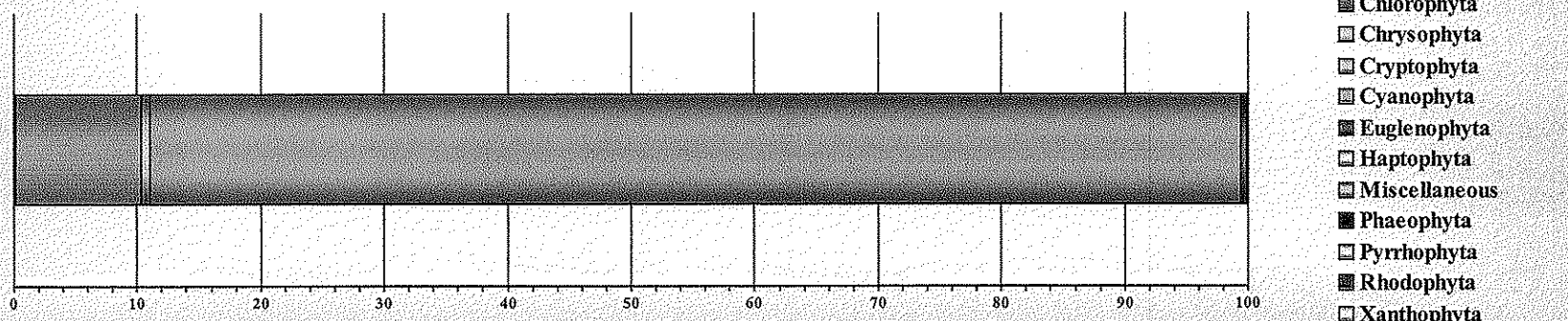
Division: Pyrrhophyta

Taxa ID	Genus	Species	Subspecies	Variety	Form	Morph	Structure	GALD µm	Count NU/ml	Relative Count	Total Biovolume µm^3 /ml	Relative Total Biovolume
6034	*Gymnodinium*	*sp. 3*	.	.	.	.	Vegetative	16.00	8.778	0.05	5,294.687	0.01
6033	*Gymnodinium*	*sp. 2*	.	.	.	.	Vegetative	20.00	13.167	0.08	17,648.956	0.05
6032	*Gymnodinium*	*sp. 1*	.	.	.	.	Vegetative	32.00	4.389	0.03	21,178.746	0.06
Summary for Division ~ Pyrrhophyta (3 detail records)							**Sum Total Pyrrhophyta**		26.334	0.16	44,122.388	0.12

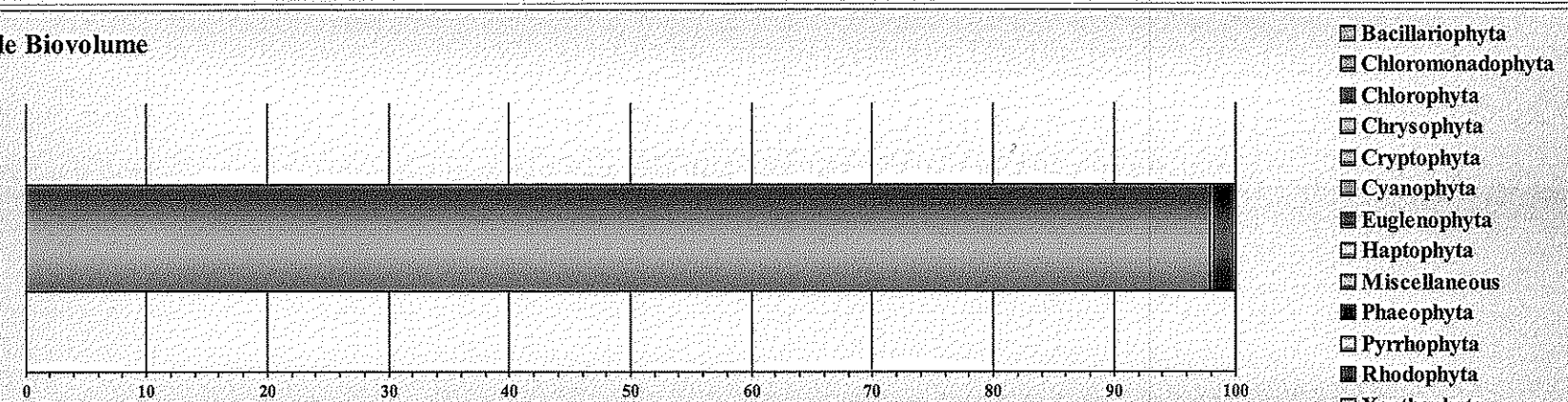
Total Sample Concentration	Total Sample Cell Concentration
16,658.206	35,705,846.010

SUMMARY GRAPHICS

Sample Concentration



Sample Biovolume



Tracking Code:	050002-000	**Sample ID:**	Phytoplankton Grab with Biovolume	**Replicate:**	1
Customer ID:	000	**Sample Date:**	12/16/2004	**Sample Level:**	Composite
Job ID:	1	**Station:**	Sample Station	**Sample Depth:**	0
System Name:	Sample Lake	**Site:**	Sample Site	**Preservative:**	Lugols
Report Notes:	Sample Report Data				

Division: Bacillariophyta

Taxa ID	Genus	Species	Subspecies	Variety	Form	Morph	Structure	GALD µm	Count NU/ml	Relative Count	Total Biovolume µm^3 /ml	Relative Total Biovolume
9123	*Nitzschia*	*palea*	.	.	.	.	Vegetative	40.00	2.726	0.28	685.196	0.71
Summary for Division ~ Bacillariophyta (1 detail record)							**Sum Total Bacillariophyta**		2.726	0.28	685.196	0.71

Division: Chlorophyta

Taxa ID	Genus	Species	Subspecies	Variety	Form	Morph	Structure	GALD µm	Count NU/ml	Relative Count	Total Biovolume µm^3 /ml	Relative Total Biovolume
2211	*Dictyosphaerium*	*pulchellum*	.	.	.	.	Vegetative	40.00	2.726	0.28	1,233.353	1.28
8041	*Monoraphidium*	*capricornutum*	.	.	.	.	Vegetative	4.00	2.726	0.28	21.295	0.02
2483	*Scenedesmus*	*bijuga*	.	.	.	.	Vegetative	12.00	2.726	0.28	154.169	0.16
Summary for Division ~ Chlorophyta (3 detail records)							**Sum Total Chlorophyta**		8.179	0.85	1,408.817	1.46

Division: Chrysophyta

Taxa ID	Genus	Species	Subspecies	Variety	Form	Morph	Structure	GALD µm	Count NU/ml	Relative Count	Total Biovolume µm^3 /ml	Relative Total Biovolume
7111	*Gonyostomum*	*semen*	.	.	.	.	Vegetative	70.00	2.726	0.28	19,984.886	20.71
1180	*Mallomonas*	*spp*	.	.	.	.	Vegetative	21.00	5.453	0.57	18,226.216	18.88
Summary for Division ~ Chrysophyta (2 detail records)							**Sum Total Chrysophyta**		8.179	0.85	38,211.101	39.59

Division: Cryptophyta

Taxa ID	Genus	Species	Subspecies	Variety	Form	Morph	Structure	GALD µm	Count NU/ml	Relative Count	Total Biovolume µm^3 /ml	Relative Total Biovolume
3015	*Cryptomonas*	*erosa*	.	.	.	.	Vegetative	14.00	84.516	8.78	21,831.106	22.62
3069	*Cryptomonas*	*rostratiformis*	.	.	.	.	Vegetative	48.00	2.726	0.28	19,733.647	20.45
Summary for Division ~ Cryptophyta (2 detail records)							**Sum Total Cryptophyta**		87.242	9.07	41,564.753	43.06

Division: Cyanophyta

Taxa ID	Genus	Species	Subspecies	Variety	Form	Morph	Structure	GALD µm	Count NU/ml	Relative Count	Total Biovolume µm^3 /ml	Relative Total Biovolume
4054	*Aphanocapsa*	*delicatissima*	.	.	.	.	Vegetative	20.00	2.726	0.28	45.680	0.05
4285	*Synechocystis*	*spp*	.	.	.	>1 um spherical	Vegetative	1.00	845.156	87.82	442.524	0.46
Summary for Division ~ Cyanophyta (2 detail records)							**Sum Total Cyanophyta**		847.882	88.10	488.203	0.51

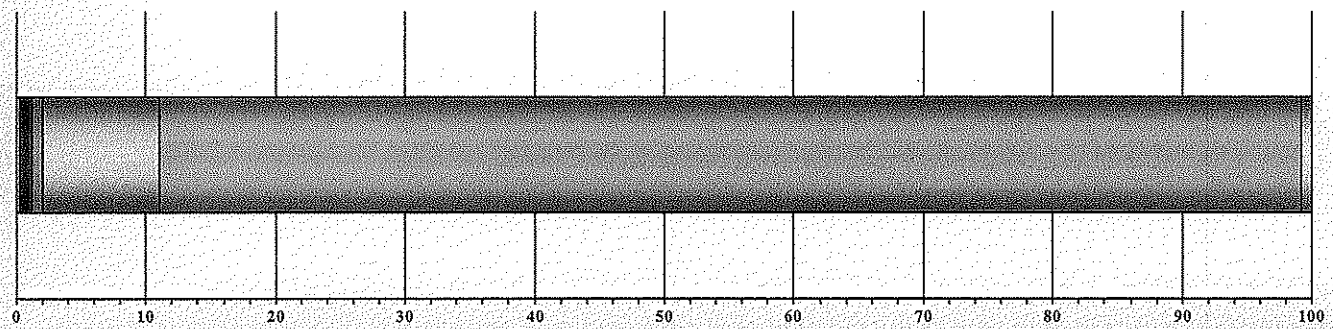
Division: Pyrrhophyta

Taxa ID	Genus	Species	Subspecies	Variety	Form	Morph	Structure	GALD µm	Count NU/ml	Relative Count	Total Biovolume µm^3 /ml	Relative Total Biovolume
6032	*Gymnodinium*	*sp. 1*	.	.	.	.	Vegetative	24.00	2.726	0.28	6,851.961	7.10
6033	*Gymnodinium*	*sp. 2*	.	.	.	.	Vegetative	20.00	5.453	0.57	7,308.758	7.57
Summary for Division ~ Pyrrhophyta (2 detail records)							**Sum Total Pyrrhophyta**		8.179	0.85	14,160.719	14.67

Total Sample Concentration	Total Sample Cell Concentration
962.387	96,518.789

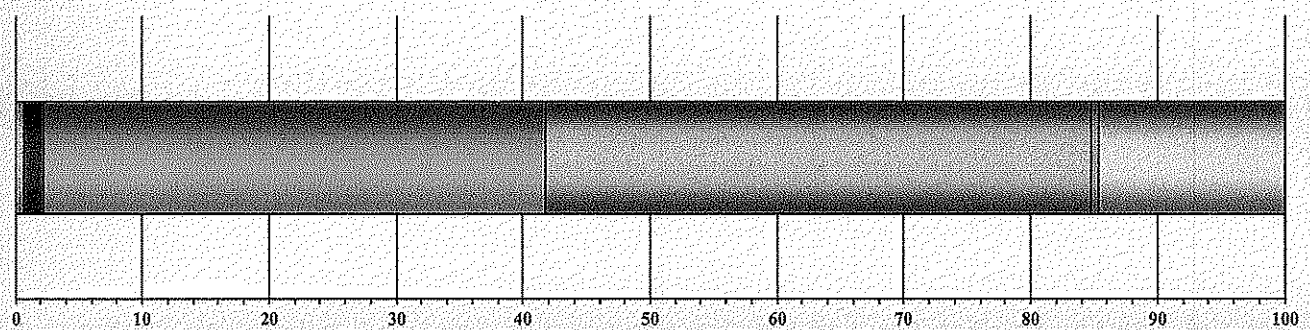
SUMMARY GRAPHICS

Sample Concentration



- Bacillariophyta
- Chloromonadophyta
- Chlorophyta
- Chrysophyta
- Cryptophyta
- Cyanophyta
- Euglenophyta
- Haptophyta
- Miscellaneous
- Phaeophyta
- Pyrrhophyta
- Rhodophyta
- Xanthophyta

Sample Biovolume



- Bacillariophyta
- Chloromonadophyta
- Chlorophyta
- Chrysophyta
- Cryptophyta
- Cyanophyta
- Euglenophyta
- Haptophyta
- Miscellaneous
- Phaeophyta
- Pyrrhophyta
- Rhodophyta
- Xanthophyta

Tracking Code:	050003-000	**Sample ID:**	Phytoplankton Grab with Biovolume	**Replicate:**	1
Customer ID:	000	**Sample Date:**	11/11/2004	**Sample Level:**	Composite
Job ID:	1	**Station:**	Sample Station	**Sample Depth:**	0
System Name:	Sample Lake	**Site:**	Sample Site	**Preservative:**	Lugols
Report Notes:	Sample Report Data				

Division: Bacillariophyta

Taxa ID	Genus	Species	Subspecies	Variety	Form	Morph	Structure	GALD µm	Count NU/ml	Relative Count	Total Biovolume µm^3 /ml	Relative Total Biovolume
1221	*Nitzschia*	*acicularis*	.	.	.	.	Vegetative	100.00	45.439	0.17	32,118.564	0.19
9506	*Synedra*	*ulna*	.	ulna	.	.	Vegetative	64.00	45.439	0.17	26,172.562	0.16
9114	*Nitzschia*	*fonticola*	.	.	.	.	Vegetative	34.00	90.877	0.34	62,866.739	0.37
Summary for Division ~ Bacillariophyta (3 detail records)							**Sum Total Bacillariophyta**		181.754	0.67	121,157.865	0.72

Division: Chlorophyta

Taxa ID	Genus	Species	Subspecies	Variety	Form	Morph	Structure	GALD µm	Count NU/ml	Relative Count	Total Biovolume µm^3 /ml	Relative Total Biovolume
8041	*Monoraphidium*	*capricornutum*	.	.	.	.	Vegetative	6.00	45.439	0.17	50.700	0.00
2340	*Mougeotia*	*spp*	.	.	.	.	Vegetative	1,100.00	90.877	0.34	7,851,204.976	46.75
2371	*Pandorina*	*morum*	.	.	.	.	Vegetative	35.00	45.439	0.17	10,141.359	0.06
1000516	*Pedinomonas*	*minutissima*	.	.	.	.	Vegetative	4.00	136.315	0.51	3,254.681	0.02
2590	*Ulothrix*	*spp*	.	.	.	.	Vegetative	1,920.00	45.439	0.17	6,851,960.706	40.80
Summary for Division ~ Chlorophyta (5 detail records)							**Sum Total Chlorophyta**		363.508	1.35	14,716,612.422	87.64

Division: Cryptophyta

Taxa ID	Genus	Species	Subspecies	Variety	Form	Morph	Structure	GALD µm	Count NU/ml	Relative Count	Total Biovolume µm^3 /ml	Relative Total Biovolume
3015	*Cryptomonas*	*erosa*	.	.	.	.	Vegetative	16.00	45.439	0.17	27,407.843	0.16
3043	*Rhodomonas*	*minuta*	.	nannoplanctica	.	.	Vegetative	8.00	45.439	0.17	1,332.324	0.01
Summary for Division ~ Cryptophyta (2 detail records)							**Sum Total Cryptophyta**		90.877	0.34	28,740.168	0.17

Division: Cyanophyta

Taxa ID	Genus	Species	Subspecies	Variety	Form	Morph	Structure	GALD µm	Count NU/ml	Relative Count	Total Biovolume µm^3 /ml	Relative Total Biovolume
4331	*Anabaena*	*macrospora*	.	.	.	.	Vegetative	480.00	21.945	0.08	135,685.168	0.81
4285	*Synechocystis*	*spp*	.	.	.	>1 um spherical	Vegetative	1.00	25,354.669	94.02	13,275.705	0.08
4421	*Lyngbya*	*subtilis*	.	.	.	.	Vegetative	280.00	45.439	0.17	34,259.802	0.20
4174	*Oscillatoria*	*tenuis*	.	.	.	.	Vegetative	1,300.00	45.439	0.17	1,670,165.420	9.95
Summary for Division ~ Cyanophyta (4 detail records)							**Sum Total Cyanophyta**		25,467.491	94.44	1,853,386.094	11.04

Division: Euglenophyta

Taxa ID	Genus	Species	Subspecies	Variety	Form	Morph	Structure	GALD µm	Count NU/ml	Relative Count	Total Biovolume µm^3 /ml	Relative Total Biovolume
5020	*Euglena*	*spp*	.	.	.	.	Vegetative	16.00	45.439	0.17	48,115.992	0.29
Summary for Division ~ Euglenophyta (1 detail record)							**Sum Total Euglenophyta**		45.439	0.17	48,115.992	0.29

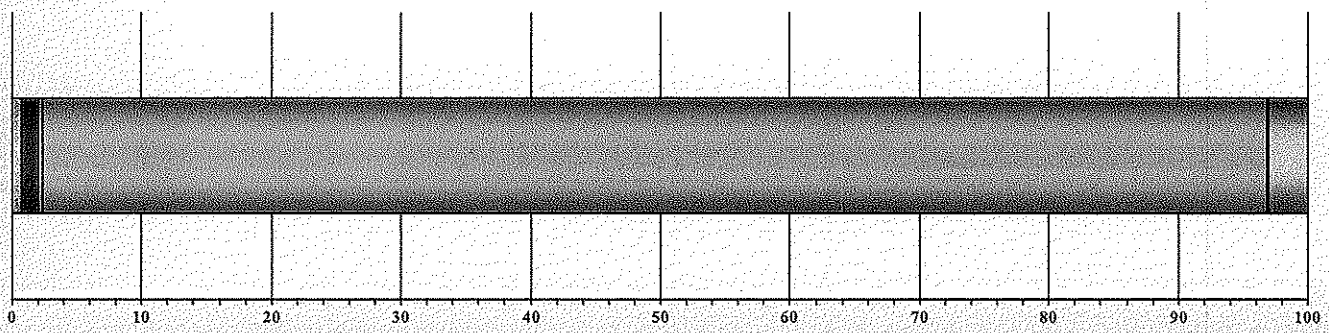
Division: Miscellaneous

Taxa ID	Genus	Species	Subspecies	Variety	Form	Morph	Structure	GALD µm	Count NU/ml	Relative Count	Total Biovolume µm^3 /ml	Relative Total Biovolume
7140	*.	*spp*	.	.	.	Microflagellate	Vegetative	4.00	817.893	3.03	24,735.607	0.15
Summary for Division ~ Miscellaneous (1 detail record)							**Sum Total Miscellaneous**		817.893	3.03	24,735.607	0.15

Total Sample Concentration	Total Sample Cell Concentration
26,966.961	16,792,748.148

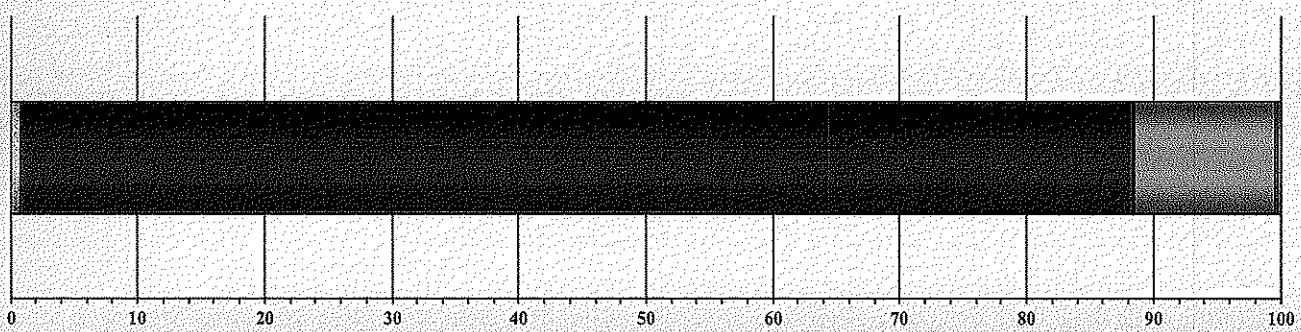
SUMMARY GRAPHICS

Sample Concentration



- Bacillariophyta
- Chloromonadophyta
- Chlorophyta
- Chrysophyta
- Cryptophyta
- Cyanophyta
- Euglenophyta
- Haptophyta
- Miscellaneous
- Phaeophyta
- Pyrrhophyta
- Rhodophyta
- Xanthophyta

Sample Biovolume



- Bacillariophyta
- Chloromonadophyta
- Chlorophyta
- Chrysophyta
- Cryptophyta
- Cyanophyta
- Euglenophyta
- Haptophyta
- Miscellaneous
- Phaeophyta
- Pyrrhophyta
- Rhodophyta
- Xanthophyta

Species List

Division: Bacillariophyta

Taxa ID	Genus	Species	Subspecies	Variety	Form	Morph	Structure	Authority
1000514	Eunotia	fallax	.	.	.	.	Vegetative	Cleve
1071	Cyclotella	sp. 1	.	.	.	.	Vegetative	(Kutzing) de Brebisson
1076	Cyclotella	meneghiniana	.	.	.	.	Vegetative	Kutzing
1210	Navicula	spp	.	.	.	.	Vegetative	Bory .
1221	Nitzschia	acicularis	.	.	.	.	Vegetative	(Kutzing) W. Smith
1298	Stephanodiscus	parvus	.	.	.	.	Vegetative	Stoermer & H†k.
1343	Amphora	pediculus	.	.	.	.	Vegetative	(Kutzing) Grunow
1523	Cyclostephanos	damasii	.	.	.	.	Vegetative	(Hustedt) Stoermer & Hakansson
9043	Fragilaria	pinnata	.	.	.	.	Vegetative	Ehrenberg
9045	Fragilaria	construens	.	.	venter	.	Vegetative	(Ehrenberg) Hustedt
9114	Nitzschia	fonticola	.	.	.	.	Vegetative	Grunow
9123	Nitzschia	palea	.	.	.	.	Vegetative	(K tz.) W. Sm.
9126	Nitzschia	subacicularis	.	.	.	.	Vegetative	Hust.
9212	Cocconeis	placentula	.	lineata	.	.	Vegetative	(Ehrenb.) Van Heurck
9458	Navicula	cf. lacunolaciniata	.	.	.	.	Vegetative	Lange-Bertalot & Bonik
9506	Synedra	ulna	.	ulna	.	.	Vegetative	(Nitzsch) Ehrenb.
9818	Stephanodiscus	medius	.	.	.	.	Vegetative	Hakansson

Division: Chlorophyta

Taxa ID	Genus	Species	Subspecies	Variety	Form	Morph	Structure	Authority
2381	Pediastrum	spp	.	.	.	.	Vegetative	Meyen
1000012	Closterium	spp	.	.	.	.	Vegetative	.
1000031	Ankistrodesmus	falcatus	.	.	.	straight	Vegetative	(Corda) Ralfs
1000516	Pedinomonas	minutissima	.	.	.	.	Vegetative	Korshikov

102793	Scenedesmus	acutus	alternans	.	.	.	Vegetative	.
102813	Scenedesmus	bijuga	alternans	.	.	.	Vegetative	(Reinsch) Hansg.
10611	Coelastrum	reticulatum	.	duplex	.	.	Vegetative	Compere;
2031	Ankistrodesmus	falcatus	.	.	.	monoraphidiod	Vegetative	(Corda) Ralfs
2035	Ankistrodesmus	convolutus	.	.	.	.	Vegetative	Corda
2080	Chlamydomonas	spp	.	.	.	.	Vegetative	Ehrenberg
2082	Chlamydomonas	globosa	.	.	.	.	Vegetative	Snow
2085	Chlamydomonas	platystigma	.	.	.	.	Vegetative	Pascher
2171	Coelastrum	microporum	.	.	.	.	Vegetative	Naeg
2173	Coelastrum	cambricum	.	.	.	.	Vegetative	Archer
2175	Coelastrum	pseudomicroporum	.	.	.	.	Vegetative	Kors
2211	Dictyosphaerium	pulchellum	.	.	.	.	Vegetative	Wood
2324	Kirchneriella	obesa	.	.	.	.	Vegetative	(W. West) Schmidle
2340	Mougeotia	spp	.	.	.	.	Vegetative	Kisselew
2363	Oocystis	parva	.	.	.	.	Vegetative	West & West
2367	Oocystis	pusilla	.	.	.	.	Vegetative	Hansgirg
2371	Pandorina	morum	.	.	.	.	Vegetative	(O. Muller) Bory De St Vincent
2483	Scenedesmus	bijuga	.	.	.	.	Vegetative	(Turpin) Lagerh.
2487	Scenedesmus	dimorphus	.	.	.	.	Vegetative	(Turpin) Kuetzing
2488	Scenedesmus	denticulatus	.	.	.	.	Vegetative	Lagerhiem
2491	Schroederia	judayi	.	.	.	.	Vegetative	G. M. Smith
2501	Selenastrum	minutum	.	.	.	.	Vegetative	(Naegeli) Collins
2554	Tetraedron	minimum	.	.	.	.	Vegetative	(Braun) Hansgirg
2561	Tetrastrum	staurogeniaeforme	.	.	.	.	Vegetative	(Schroeder) Lemm.
2590	Ulothrix	spp	.	.	.	.	Vegetative	Kuetzing
2683	*Chlorococcaceae	spp	.	.	.	2-9.9 um spherical	Vegetative	N/A
2684	*Chlorococcaceae	spp	.	.	.	> 10 um spherical	Vegetative	N/A

2687	*Chlorococcaceae	spp	.	.	.	> 1 um ovoid	Vegetative	(Brandt) Beijerinck
2830	Nephrocytium	spp	.	.	.	.	Vegetative	Naegel
2840	Lobomonas	spp	.	.	.	.	Vegetative	Dangeard
2861	Monomastix	astigmata	.	.	.	.	Vegetative	Skuja
2884	Scenedesmus	quadricauda	.	.	.	.	Vegetative	(Turpin) Breb.
2892	Staurastrum	paradoxum	.	.	.	.	Vegetative	Meyer
2911	Stichococcus	bacillaris	.	.	.	.	Vegetative	Nageli
2950	Spirogyra	spp	.	.	.	.	Vegetative	Skuja
8041	Monoraphidium	capricornutum	.	.	.	.	Vegetative	(Printz) Nygaard
8226	Scenedesmus	intermedius	.	.	.	.	Vegetative	Chodat
8303	Scenedesmus	opoliensis	.	carinatus	.	.	Vegetative	Lemmermann
8308	Scenedesmus	serratus	.	.	.	.	Vegetative	(Corda) Bohlin
8332	Tetraedron	muticum	.	.	.	.	Vegetative	(A. Braun) Hansgirg

Division: Chrysophyta

Taxa ID	Genus	Species	Subspecies	Variety	Form	Morph	Structure	Authority
1180	Mallomonas	spp	.	.	.	.	Vegetative	Perty
1183	Mallomonas	akrokomas	.	.	.	.	Vegetative	Smith
1323	Synura	spp	.	.	.	.	Vegetative	Ehrenberg
1570	Ochromonas	spp	.	.	.	.	Vegetative	Wyssotzki
1731	Erkenia	subaequiciliata	.	.	.	.	Vegetative	Skuja
7111	Gonyostomum	semen	.	.	.	.	Vegetative	.

Division: Cryptophyta

Taxa ID	Genus	Species	Subspecies	Variety	Form	Morph	Structure	Authority
3015	Cryptomonas	erosa	.	.	.	.	Vegetative	Ehrenberg .
3041	Rhodomonas	minuta	.	.	.	.	Vegetative	Skuja
3043	Rhodomonas	minuta	.	nannoplanctica	.	.	Vegetative	Skuja

3069	Cryptomonas	rostratiformis	.	.	.	.	Vegetative	Skuja

Division: Cyanophyta

Taxa ID	Genus	Species	Subspecies	Variety	Form	Morph	Structure	Authority
107576	Lyngbya	lagerheimia	.	minor	.	.	Vegetative	(Moebius) Gomont
4041	Aphanizomenon	flos-aquae	.	.	.	.	Vegetative	(L.) Ralfs
4051	Aphanocapsa	elachista	.	.	.	.	Vegetative	West & West
4054	Aphanocapsa	delicatissima	.	.	.	.	Vegetative	West & West
4062	Aphanothece	nidulans	.	.	.	.	Vegetative	P. Richter
4152	Lyngbya	contorta	.	.	.	.	Vegetative	Lemm
4172	Oscillatoria	limnetica	.	.	.	.	Vegetative	Lemmermann
4174	Oscillatoria	tenuis	.	.	.	.	Vegetative	Agardh
4183	Oscillatoria	agardhii	.	.	.	.	Vegetative	Gomont
4285	Synechocystis	spp	.	.	.	>1 um spherical	Vegetative	N/A
4321	Synechococcus	elongatus	.	.	.	.	Vegetative	Nageli
4331	Anabaena	macrospora	.	.	.	.	Vegetative	Klebahn 1895
4421	Lyngbya	subtilis	.	.	.	.	Vegetative	West & West .
4460	Pseudanabaena	spp	.	.	.	.	Vegetative	Lauterborn

Division: Euglenophyta

Taxa ID	Genus	Species	Subspecies	Variety	Form	Morph	Structure	Authority
118570	Lepocinclis	glabra	.	.	.	.	Vegetative	.
5020	Euglena	spp	.	.	.	.	Vegetative	Ehrenberg
5023	Euglena	acus	.	.	.	.	Vegetative	Ehrenberg
5030	Phacus	spp	.	.	.	.	Vegetative	Dujardin .
5036	Phacus	helikoides	.	.	.	.	Vegetative	Pochmann

Division: Miscellaneous

Taxa ID	Genus	Species	Subspecies	Variety	Form	Morph	Structure	Authority
7140	*.	spp	.	.	.	Microflagellate	Vegetative	N/A

Division: Pyrrhophyta

Taxa ID	Genus	Species	Subspecies	Variety	Form	Morph	Structure	Authority
6032	Gymnodinium	sp. 1	.	.	.	.	Vegetative	Stein
6033	Gymnodinium	sp. 2	.	.	.	.	Vegetative	Stein
6034	Gymnodinium	sp. 3	.	.	.	.	Vegetative	Stein
6040	Peridinium	spp	.	.	.	.	Vegetative	Ehrenberg
6044	Peridinium	umbonatum	.	.	.	.	Vegetative	Stein