STATE OF TENNESSEE

DEPARTMENT OF ENVIRONMENT AND CONSERVATION

Division of Water Pollution Control

Quality System Standard Operating Procedure

for

PERIPHYTON STREAM SURVEYS

January 2010

This SOP is an intra-departmental document intended to govern the internal management of the Tennessee Department of Environment and Conservation and to meet requirements of the U.S. Environmental Protection Agency for a quality system. It is not intended to affect rights, privileges, or procedures available to the public.

DIVISION OF WATER POLLUTION CONTROL
QUALITY SYSTEMS STANDARD OPERATING PROCEDURES
FOR PERIPHYTON STREAM SURVEYS

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DIVISION OF WATER POLLUTION CONTROL

QUALITY SYSTEM STANDARD OPERATING PROCEDURE FOR PERIPHYTON STREAM SURVEYS

TITLE AND APPROVAL PAGE

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PLAN COVERAGE	General instructions for periphyton stream surveys in Tennessee

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REVISIONS AND ANNUAL REVIEW PROCEDURE: QS-SOP FOR PERIPHYTON STREAM SURVEYS

1. This document shall be reviewed annually to reconfirm the suitability and effectiveness of the program components described in this document.

2. A report of the evaluation of effectiveness of this document shall be developed at the time of review and submitted to appropriate stakeholders. Peer Reviews shall be conducted, if necessary and appropriate. It shall be reconfirmed that the document is suitable and effective. It shall include, if necessary, clarification of roles and responsibilities, response to problem areas and acknowledgement of successes. Progress toward meeting TDEC–BOE mission, program goals and objectives shall be documented. Plans shall be made for the upcoming cycle and communicated to appropriate stakeholders.

3. The record identified as "Revisions" shall be used to document all changes.

4. A copy of any document revisions made during the year shall be sent to all appropriate stakeholders. A report shall be made to the Assistant Commissioner and Quality Assurance Manager of any changes that occur. Other stakeholders shall be notified, as appropriate and documented on the "Document Distribution" list.

NOTICE OF REVISION(S) RECORD

Date	Specific Section or Page	Revision Type (major or minor)	Revision Description

EVALUATION PROCEDURE:
QS-SOP FOR PERIPHYTON STREAM SURVEYS

As this document is used, needed changes or improvements will be apparent. Specific recommendations for improvements or changes are solicited as well as information concerning typographical or formatting errors.

1. Copy this page and complete all questions. Electronic versions of this are encouraged especially if comments are significant.

2. Send specific recommendations for improvements or changes, along with the following information, to:

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E-mail Address _____
Telephone Number _____
Document Effective Date _____
Section(s) and Page Number(s) to which your comments apply _____
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QS-SOP DOCUMENT DISTRIBUTION LIST

Copies of this document were distributed to the following individuals in TDEC and TDH

Additional copies were distributed to non-TDEC agencies and individuals upon request (including other state and federal agencies, consultants, universities etc.). An updated distribution list is maintained in the Planning and Standards Section.

The system for document distribution is described in TDEC-BOE Quality Manual, Chapters 5 and 10.

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PREFACE

The U.S. EPA requires that a centrally planned, directed and coordinated quality assurance and quality control program be applied to efforts supported by them through grants, contracts or other formalized agreements. This includes the implementation of a Quality Management Plan as written by the contract holder with Data Quality Objectives (DQOs) set in Quality Assurance Project Plans (QAPPs) for specific projects. The organization may elect to support portions of the QAPP through technical or administrative standard operating procedures (SOPs), as specified by the quality system. As a contract holder and through memoranda of agreement, the Tennessee Department of Environment and Conservation is required to maintain such a system.

This quality system technical Standard Operating Procedure (QS-SOP) was prepared, reviewed and distributed in accordance with TDEC's Quality Management Plan and other quality system documents in response to U.S. EPA's requirements for a Quality Management Program. QS-SOPs are integral parts of successful quality systems as they provide staff with the information to perform a job properly and facilitate consistency in the quality and integrity of the process.

This QS-SOP is specific to the Division of Water Pollution Control and is intended to assist the division in maintaining their quality control and quality assurance processes and to ensure compliance with government regulations. It provides specific operational direction for the division's Quality Assurance Project Plan for Periphyton Stream Surveys.

I. PROCEDURES

I.A SCOPE, APPLICABILITY AND REGULATORY REQUIREMENTS

The purpose of this Quality Systems Standard Operating Procedure (QS-SOP) is to support the Quality Assurance Program. The document provides a consolidated reference document for use in training and orientation of employees. This guide will also be a reference tool for more experienced employees. It establishes an approach that can be recommended to sister agencies that monitor Tennessee water or stipulated to members of the regulated community given monitoring requirements in receiving streams. This SOP describes the periphyton stream survey process and will delineate all steps in the process, including habitat assessments, field collections, sample analysis, data reduction and reporting. This SOP is only intended to describe routine conditions encountered during a periphyton stream survey.

Federal Statutory Authority

Federal Water Pollution Control Act (amended through P.L. 106-308, October 13,2000) as Amended by the Clean Water Act of 1977 enacted by Public Law 92-500, October 18, 1972, 86 Stat. 816; 33 U.S.C. 1251 et. seq.
Title III, Sec. 302: Water Quality Related Effluent Limitations
Title III, Sec. 303: Water Quality Standards and Implementation Plans
Title III, Sec. 304: Information and Guidelines
Title III, Sec. 305: Water Quality Inventory

Tennessee Statutory Authority

Tennessee Water Quality Control Act of 1977 (Acts 1971, ch. 164, § 1; 1977 ch. 366, § 1; T.C.A., § 69-3-101 et seq.

Tennessee Regulatory Authority

General Water Quality Criteria and the Antidegradation Statement: Rule 1200-4-3
(specifically 1200-4-3-.03(3) j: Biological Integrity and 1200-4-3-.06 Tennessee Antidegradation Statement)
Use Classification for Surface Waters: Rule 1200-4-4

I.B METHOD SUMMARY

This document describes procedures for performing periphyton surveys approved by the Division of Water Pollution Control for assessing biological integrity of streams. The entire procedure is described, including protocols for sample collection, habitat assessment, sample analysis, data reduction and reporting.

Due to the sedentary nature of periphyton, the community composition and biomass are sensitive to changes in water quality. A diverse community of periphyton can be found in healthy streams. Nuisance blooms are usually symptoms of a system stressed by factors such as excessive nutrients, elevated temperatures, or stagnant conditions. Therefore, the division will use periphyton as secondary biological indicators of impairment in streams. The primary biological indicators are macroinvertebrates.

Two periphyton sampling protocols are described in this document. Both will be performed every time periphyton sampling is indicated. The first, Rapid Periphyton Survey (RPS), is an estimate of algal biomass found in the stream. The second, more intensive protocol involves collecting a sample of periphyton and analyzing the periphyton community found there. Both sampling protocols will be carried out in monitoring stations where nutrient impairment is suspected or known, as well as ecoregion reference streams.

Habitat assessments (high gradient and low gradient) are also described in this document. Habitat assessments are to be conducted in conjunction with all types of biological surveys since habitat is often a limiting factor to the complexity of the benthic community. By following this assessment procedure, habitat can either be confirmed or eliminated as a cause of stress to the periphyton community.

I.C DEFINITIONS AND ACRONYMS

Ambient Monitoring: Routine sampling and evaluation of receiving waters not necessarily associated with periodic disturbance.

Benthic Community: Plants and animals living on the bottom of the stream.

Ecoregion: A relatively homogenous area defined by similarity of climate, landform, soil, potential natural vegetation, hydrology, and other ecologically relevant variables. There are eight (Level III) ecoregions in Tennessee.

Ecological Subregion (or subecoregion): A smaller area that has been delineated within an ecoregion that has even more homogenous characteristics than does the original ecoregion. There are 31 (Level IV) ecological subregions in Tennessee.

Ecoregion Reference: Least impacted waters within an ecoregion that have been monitored to establish a baseline to which alterations of other waters can be compared.

Frustule: The silicate cell wall of a diatom. The frustule is composed of two valves.

Habitat: The instream and riparian features that influence the structure and function of the aquatic community in a stream.

Macroalgae: Long filamentous strands of algae such as *Cladophora* or *Spirogyra* spp.

Microalgae: Primarily single celled algae which coat the substrate and are generally composed of diatoms and soft algae such as blue-green algae

Moss: Member of the division Bryophyta

Periphyton: Algae attached to submerged substrate in aquatic environments.

Reference database: Biological and chemical data from ecoregion reference sites.

Riparian Zone: An area that borders a waterbody (approximately 18 meters wide).

Valve: Half of a frustule.

Watershed: The area that drains to a particular body of water or common point.

Acronyms

CGR	*Cymbella* Group Richness
D.O.	Dissolved Oxygen
D/S	Downstream
EFO	Environmental Field Office
ES	Environmental Specialist
FGR	*Fragilaria* Group Richness
GPS	Global Positioning System
H’	Shannon Diversity Index
KDBI	Kentucky Diatom Bioassessment Index
KPTI	Kentucky Pollution Tolerance Index
LDB	Left Descending Bank
MPS	Multi-habitat Periphyton Sample
MT	Mountain Region
MVIR	Mississippi Valley-Interior Region
% NNS	Percent *Navicula*, *Nitzschia*, and *Surirella*
PAS	Planning and Standards Section
PN	Pennyroyal Region
QA/QC	Quality Assurance/Quality Control
RDB	Right Descending Bank
RPS	Rapid Periphyton Survey
SOP	Standard Operating Procedure
TDEC	Tennessee Department of Environment and Conservation
TMDL	Total Maximum Daily Loading
TNDT	Total Number of Diatom Taxa
TOPO	Topographic Map
TR	Taxa Richness
U/S	Upstream
WPC	Water Pollution Control

I.D HEALTH AND SAFETY WARNINGS

(Adopted from Klemm et al., 1990)

1. Know how to swim and/or use personal flotation devices when entering the water.

2. Always wear a belt with chest waders to prevent them from filling with water in case of a fall. If it is necessary to wade in high velocity and high flow streams it is advisable to wear a life jacket.

3. Follow Tennessee boating laws and regulation. Information is available through the Tennessee Wildlife Resources Agency (TWRA). Life jackets are required when operating a boat.

4. Be vigilant, especially in turbid streams, to avoid broken glass, beaver traps or other sharp objects that may lie out of sight on the bottom.

5. Keep first aid supplies in the EFO and in the field at all times. Training in basic first aid and cardio-pulmonary resuscitation is strongly recommended.

6. Any person allergic to bee stings or other insect bites should have needed medications in the event of an allergic reaction and instruct others in the team on how to use the allergy kit. Also, be aware of and alert to the potential presence of venomous snakes.

7. Always handle formaldehyde under a properly installed and operating fume hood. Check to be certain the fume hood is functioning properly. Always wear personal protective equipment (gloves, safety glasses, and lab coat) when handling preservatives. Know the location and how to use eyewash and shower stations.

8. It is recommended that communication equipment be taken in the field in case of an emergency.

9. Keep an employee file in the field office that contains emergency contacts and physician's name for each employee. Carry a list of emergency contact numbers to the sample area. Know the location and emergency contact numbers of hospitals and law enforcement stations in the area.

10. Consider all surface waters a potential health hazard due to toxic substances or pathogens. Minimize exposure as much as possible and avoid splashing. Wearing powder-free gloves limits exposure to potential health hazards. Clean exposed body parts (face, hands, and arms) immediately after contact with these waters. Carry phosphate-free soap and an adequate supply of clean water, disinfectant wipes, and/or waterless sanitizer. Thoroughly wash exposed areas with soap and water as soon as possible.

It is recommended that powder-free gloves be worn when handling samples. In waters known or suspected to have high pathogen levels the sampler may choose to wear shoulder length gloves.

11. If working in water known or suspected to contain human wastes, EPA recommends immunization against tetanus, hepatitis, and typhoid fever (Table 1). The Tennessee Department of Health has reviewed studies that indicate a minuscule risk of contracting Hepatitis without coming in direct contact with an infected person and do not believe vaccinations are warranted. Beginning August 2002, the TDH has denied WPC request for such vaccinations. However, this does not preclude employees from contacting their physician and requesting vaccinations they believe are appropriate.

Table 1: EPA Recommended Vaccinations

Vaccination	No. of shots	Interval	Booster
Hepatitis B	3	0, 1, 6 months	NA
Tetanus	1	NA	10 years
Polio	1, if childhood series completed	NA	20 years
Typhoid	2	1 month	3 years

12. Try to avoid working alone in the field. If working alone, make sure your supervisor or their designee knows where you are and when you are expected to return. Check in periodically.

13. Material Safety Data Sheets (MSDS) are available for all preservatives and other hazardous chemicals. Everyone working with these agents or handling preserved bottles must be familiar with the location and contents of the MSDS. Notify supervisor if MSDS sheets cannot be located.

14. Be aware of potentially volatile situations. If possible, obtain permission from landowners before crossing private property. Have business cards available to leave at residences when appropriate.

15. Check to make sure lids are tightly fastened.

16. When traveling in a state vehicle always wear a seat belt and follow all Tennessee Department of Safety and Motor Vehicle Management rules.

18. In the event of a life-threatening emergency, go to the nearest hospital. Call for emergency assistance if moving the injured person is likely to inflict further injury. If a non-life threatening injury occurs on the job, seek medical assistance from the authorized state worker's compensation network. A current list of providers may be found on the State Treasurer's homepage under Workers Compensation, Provider Directory at www.treasury.tn.gov. Always complete and file an accident report if medical assistance is provided for a work related injury.

I.E CAUTIONS

1. Avoid cross contamination of samples. Use new sample bottles whenever possible, otherwise thoroughly rinse bottles and inspect before use.

2. Avoid sampling bias by following these procedures exactly. Take care not to over sample or under sample.

3. Use the standardized station ID naming protocol for all samples. Check the Water Quality Database for consistency in naming existing streams. Make sure the station ID is included on all paperwork associated with the sample.

4. To avoid errors, it is recommended to calibrate all meters at the beginning of each day (unless overnight travel is required). The meters should minimally be calibrated once a week. Perform a drift check at the end of each day (or on return to office if overnight sampling). If the meter calibration is off by more than 0.2 units for pH, temperature, or D.O. when measured in mg/L, by more than 10% for conductivity, or 10% D.O. when measured in % saturation, precede all readings between the initial calibration and the drift check with an N (questionable data) on the Rapid Periphyton Survey sheet (RPS) and on any Chemical Request Forms turned in at the TDH Environmental lab or discard the data. If sample request forms have already been submitted, notify the Planning and Standards Section of questionable readings in writing (e-mail or fax).

5. Record all time in a 24-hour (military) clock format

6. Write all dates in mm/dd/yy or mm/dd/yyyy format. (For example, March 2, 2003 would be 03/02/03 or 03/02/2003.)

7. Record all distance measurements except flow in meters.

8. Express temperature readings in degrees centigrade.

9. When performing a rapid periphyton survey, always measure canopy cover using a densiometer at the mid-point of each transect.

10. If an error is made in any documentation, draw a single line through the error, so that it is readable and write the correction above. Date and initial the correction. Do not white out or place several lines through errors.

11. Collect samples in opaque brown bottles and avoid exposure to light.

12. Make sure a minimum of 40 mL is collected for each sample.

I.F INTERFERENCES

1. Document all deviations from protocol.

2. Avoid sampling in flooded conditions or immediately after a flood. After the water level has receded to base flow, wait at least one week to sample. Avoid scoured areas.

3. Do not sample if stream is reduced to isolated pools. If stream channel naturally goes dry, only sample if there has been flow for longer than 30 days.

4. Sampling should be conducted between March and November (preferably between April and October) when periphyton growth is highest and rainfall runoff is generally lower. High flow levels and scouring may be greater in the winter and spring.

5. Flag dissolved oxygen, pH, temperature and conductivity readings with an N (Questionable data) if post-trip drift checks show meter calibrations to be off by more than 0.2 units (or 10% for conductivity) or discard the data.

6. Sampling stations should be located in areas where the periphyton community is not influenced by atypical conditions, such as those created by bridges or dams unless judging the effects of atypical conditions is a component of the study objectives.

7. Avoid areas of deep canopy unless entire reach is shaded.

I.G PERSONNEL QUALIFICATIONS

Tennessee Civil Service Titles: Biologist, Environmental Specialist, Environmental Protection Specialist, Environmental Program Manager, Environmental Field Office Manager, or trained co-op/intern (state employees only). For the purpose of this report, all position titles will be referred to as biologist.

Minimum Education Requirements: B.S. in a biological science. Coursework in stream ecology and/or phycology is desirable. A graduate level degree in phycology, aquatic biology, stream ecology or similar field is preferable.

Minimum experience: one year (specific class-work involving biological stream surveys can be substituted for experience).

Expertise: Computation of basic statistics, use of standard water quality monitoring meters, habitat evaluations and general water quality assessments, ability to recognize macroalgae and microalgae.

Training: Protocols outlined in this SOP
- Quality System Requirements
- Quality Assurance Project Plan

I.H. EQUIPMENT AND SUPPLIES

Prior to any sampling trip, gather and inspect all necessary gear. Replace or repair any damaged equipment. Calibrate all meters the morning of the sampling trip with a drift check at the end of the day (or the end of the trip for overnight stays). Upon return from a trip, take care of any equipment repairs or replacements immediately. Necessary equipment will vary per project, but the following is a standardized list.

Field Equipment

Waders
External sample tags
Habitat Assessment Sheet (High gradient for riffles, Low gradient for glide-pool)
Biological Analysis Request Sheet (for Chain of Custody and/or samples sent to lab)
Topographic maps (USGS quadrangle maps) may also be referred to as topos or quads.
Tennessee Atlas and Gazetteer
Calibrated GPS unit
Calibrated Dissolved Oxygen meter and replacement membrane kit
Calibrated pH meter
Calibrated conductivity meter
Calibrated temperature meter or thermometer in ^{o}C
Spare batteries for all meters and for camera
Camera (preferably digital) with memory cards or film for documentation of potential pollution sources and waterbody conditions
Magnifying lens
Waterproof marking pens (Sharpies), pencils and black ballpoint ink pens (not roller-ball)
Flashlights
Duct Tape
First Aid Kit
Watch
Spherical densiometer (for canopy measurements)
Map Wheel (for calculating stream miles) if station ID is to be assigned in the field
Disposable pipettes (single squeeze approx 2 mL)
Preservative (buffered formalin)
500 mL wide mouth sample jar (approx. 9-cm inner diameter), marked at the 100 mL fill point
Scissors or knife
125 mL opaque brown plastic wide-mouth sample bottle to hold final sample
Rapid Periphyton Survey Board
Rapid Periphyton Survey Data Sheet
Small ruler

Additional Items Needed if Sampling from Sediment/Sand.

Sediment core sampler with plunger
- 60 mL plastic syringe (cut off at end)
- 3 cm depth (approx. 15-17 mL) marked on side or syringe (tape or permanent marker)

Large metal kitchen spatula
Tablespoon size measuring spoon

Laboratory Equipment

The following equipment is needed to perform sample analysis:

Tissue homogenizer or blender
Calibrated (known volume) counting chamber such as a Nannoplankton chamber or a Palmer-Maloney Counting Cell
Pipettes
Compound microscope
Beaker
Naphrax or other high refractive index medium
Microscope slides
Slide coverslips
Hot plate and nail polish (to seal edges of cover slips to slides where Naphrax was used
Bench Sheet
Slide Storage Box
Sodium bicarbonate
37% formaldehyde (formalin)
pH meter
Small containers
Balance
Nitric or sulfuric acid
Fume hood
Distilled water
Counter (for tallying taxa)

Sample Container Acquisition

Sample containers are to be obtained through the Tennessee Department of Health Environmental laboratory.

It is recommended that sample containers be requested at least two weeks prior to the anticipated date they need (preferably one month).

The 125 mL opaque amber plastic wide-mouth sample bottles used for the final periphyton collection will be available directly from Laboratory Services:

Contact: Dr. Bob Read
(615) 262-6302
bob.read@state.tn.us

I.I PROCEDURES

Protocol A - Selection of Survey Type and Station Location

Biologist or Environmental Specialist from EFO
Central Office Coordinator

1. Determine biological sampling needs.

   The central office will coordinate biological sampling needs with the environmental field offices. The location and type of biological assessments are included in the annual water quality monitoring work plan. The most common reason for periphyton sampling will be suspicion of nutrient enrichment and reference stream monitoring; however, some studies will require periphyton sampling for other reasons. It is highly recommended that nutrient samples (Nitrate + Nitrite, Total Phosphorus, Total Kjeldahl Nitrogen, Total Organic Carbon, and Ammonia) be collected at the same time. **Sampling should be conducted between March and November (preferably between April and October).**

2. Select sites.

   Site selection is dependent on the study objectives. After determining the specific objectives of the study and clearly defining what information is needed, select sampling sites on specific reaches of the stream. Reconnaissance of the waterway is very important. Note possible sources of pollution, access points, substrate types, flow characteristics, and other physical characteristics that will need to be considered in selecting the sampling sites. Although the number and location of sampling stations will vary with each individual study, the following basic rules should be applied:

   a. For **watershed screenings**, locate sites near the mouth of each tributary. If impairment is observed, locate additional sites upstream of the impaired stream reach and try to define how far the impairment extends.

   b. For monitoring **point source** pollution, establish a station downstream of the source of pollution in the stream after mixing has occurred. If complete mixing of the discharge does not occur immediately, left bank, mid-channel and right bank stations may need to be established to determine the extent of possible impact. Establish stations at various distances downstream from the discharge. Space the collecting stations exponentially farther apart going downstream from the pollution source to determine the extent of the recovery zone.

   c. For **site specific** sampling, locations immediately above, or below the confluence of two streams, or immediately below point/nonpoint source discharges should be avoided if mixing does not immediately occur. Unless the stream is extremely small or extremely turbulent, an in-flow will usually hug the stream bank with little lateral mixing for some distance. This may result in two very different biological populations and an inaccurate

assessment of stream conditions. This can be avoided by sampling after mixing has occurred.

d. All sampling stations under comparison during a study should have **similar habitat** unless the object of the study is to determine the effects of habitat degradation.

e. Sampling stations for periphyton should be located within the same reach (200 meters or yards) of where sampling for **chemical and physical parameters** will be located. If the periphyton are collected more than 200 meters from the chemical sampling, consider it a separate station and assign it a different station ID number, unless there are no tributaries, discharges or other streamside activities that would influence the stream between sampling points.

f. Sampling stations should be located in areas where the periphyton community is not influenced by **atypical conditions**, such as those created by bridges or dams unless judging the effects of atypical conditions is a component of the study objectives.

Protocol B – Assigning Station Identification Numbers

Sampler

Assign station numbers to each site using the following protocol. The station number is used to identify the sample and must be included on all associated paperwork, results, tags, etc. This number is to be used to identify this site every time it is sampled (benthic, fish, algae, bacteria, or chemical). If new stations are set up that will have chemical or bacteriological monitoring, send the station information to the Planning and Standards Section as soon as the location is finalized and before results are received. (It is usually a least a month between sample collection and receipt of results.) Minimally, station information should include station ID, latitude and longitude (in decimal degrees), HUC, ecoregion, stream order and specific location information (such as road crossing) that can be located on a map. Contact PAS if assistance is needed to assign station numbers. Continue to use the established naming protocol for NPDES effluent samples.

It is very important that station IDs are assigned consistently with the same location always assigned the same ID irregardless of the sample collection type, purpose, samplers or year. It is also important that river miles used in the station ID are measured as accurately as possible and correspond to the latitude and longitude for easy comparison between multiple stations on the same waterbody. The official stream name is the one found on the USGS 7.5 minute topographic map. Do not use other sources such as a gazetteer, TDOT bridge signs or local names, which may differ.

1. Before assigning a new station number, check the "current stations" table in the Water Quality Database to make sure a number has not already been assigned to that site. Even if the site has not been collected before by the EFO, a station ID may have already been assigned based on other agency data. Do not assume that a station does not exist because it has not been collected by the EFO. Contact the Planning and Standards Section if there is any question.

   If the station is not in the database, coordinate the naming of the station with staff members who may be collecting chemical and/or macroinvertebrate samples at the same site. Stations collected within 200 meters (yards) of each other are considered the same site and should be assigned the same station ID. If it is necessary to go more than 200 meters for chemical sampling or to find a suitable macroinvertebrate habitat, the samples can be assigned the same station ID provided there are no tributaries or potential pollution sources between the chemical and biological sample points. If there are tributaries or potential pollution sources, considerate it a separate station and assign it a different station ID.

The only exception to the following naming scheme is sites that have been designated as Ecoregion reference sites. These sites are always identified with either an ECO or FECO (headwater stream) designation no matter what the purpose of sampling. If new ecoregion reference sites are added, contact Planning and Standards (PAS) to determine the appropriate station number.

2. If a number does not already exist for the site, create a 12-character or less identification number. All letters in the station name are capitalized. Do not use more than 12 characters including the decimal.

   a. The first five digits will be the first five letters of the stream/river name (capitalized). If the stream/river name has more than one word, use the first letter of each word finishing out the five letters with the last word. For example, South Fork Forked Deer River would be SFFDE. If the creek name has fewer than five letters use the entire name. Do not use the words River, Creek, or Branch. (Fork is only used if the stream is also designated river, creek, branch etc.) For example, Dry Fork would be DRY but Dry Fork Creek would be DFORK.

   b. The next five characters designate the river mile. This will be written as three whole numbers, a decimal and a tenth space. For example, river mile 1.2 would be written as 001.2. Do not add zeros to make a short stream name longer. It is very important that the river mile be determined as accurately as possible. Ideally, this would be done by GIS. It can also be done using a topo map and a map wheel. The river mile is always measured upstream from the confluence with the main stem. If there are other stations located on the same stream, make sure that the assigned river miles are appropriately upstream or downstream of existing stations. If errors are discovered, contact PAS to have the station re-assigned.

   c. The last two characters designate the county or state, if not in Tennessee. Use the county and state identification table in Appendix A to determine the appropriate county or state abbreviation. The county or state is expressed with two letters; do not use the numeric state code. If the station is in another state, add an underscore _ before the two letter state abbreviation.

      Example 1: A station located at river mile 1.5 on Puncheoncamp Creek in Greene County would be PUNCH001.5GE.

      Example 2: A station located at river mile 25 on the North Fork Forked Deer River in Gibson County would be NFFDE025.0GI.

      Example 3. A station that is located in Kentucky at river mile 15.2 of Spring Creek would be SPRIN015.2_KY.

3. Unnamed Streams/Tributaries

Check a 24k scale topo map (hardcopy or GIS layer) to see if the unnamed stream is within a named geographical feature such as a Cove, Hollow, Gulf, Gulch, or Valley.

a. For streams that are not within a named geographical feature:

(1) Use the first five letters of the receiving stream the tributary enters.

(2) Use 1T for the first unnamed tributary station sampled, 2T for the second unnamed tributary sampled etc.

(3) Use a 3-character stream mile including one whole number, the decimal and a tenth space. For example river mile 1.2 would be written as 1.2.

(4) Use the two letter county or state abbreviation from Appendix A. If the station is in another state, add an underscore _ before the two letter state abbreviation.

Example 1: A station located at river mile 0.2 on an unnamed tributary that entered the North Fork Forked Deer River in Gibson County would be NFFDE1T0.2GI.

Example 2: A second station located at mile 5.5 on the same unnamed tributary would be NFFDE1T5.5GI.

Example 3: A site at river mile 8.5 on a different unnamed tributary to the North Fork Forked Deer would be NFFDE2T8.5GI.

(5) When naming an unnamed tributary to an unnamed tributary, use the first three letters of the main stem followed by the 1T1T, the river mile and the county. For example, a station at river mile 0.5 on an unnamed tributary to an unnamed tributary to Turkey Creek in Gibson County would be called TUR1T1T0.5GI.

b. For streams that are within a named geographical feature:

(1) The first five digits will be the first five letters of the name of the geographical feature (capitalized). If the feature name has more than one word, use the first letter of each word finishing out the five letters with the last word. Do not use the words Cove, Hollow, Gulch, Gulf, or Valley. If the feature name has fewer than five letters use the entire name.

(2) Add underscore_G to indicate that the station is named after a geographical feature and not a named stream. Streams with "_G" will be the main branch running through the feature. Use 1G for the first unnamed tributary that flows into the main

branch of the feature, 2G for the second unnamed tributary that flows into the feature, etc.

(3) The next three characters designate the miles upstream from the nearest named stream or waterbody. This will be written as one whole number, a decimal and a tenth space. For example, river mile 1.2 would be written as 1.2. If the stream is an unnamed tributary to the main branch (_G streams), the miles will be measured upstream from the main branch instead of the nearest named stream or waterbody.

(4) Use the two letter county or state abbreviation from Appendix A. If the station is in another state, add an underscore _ before the two letter state abbreviation.

Example 1: A station that is in Shingle Mill Hollow in Marion County and is 0.3 miles upstream from Nickajack Reservoir, which is the closest named waterbody would be SMILL_G0.3MI.

Example 2: A station that is located on an unnamed main branch in Cave Cove in Marion County that is 0.4 miles upstream of the nearest named stream would be CAVE_G0.4MI.

Example 3: A station on an unnamed tributary that is 0.2 miles upstream of the main branch in Example 2 would be CAVE1G0.2MI.

4. Wetlands

a. For named wetlands

(1) Use the first five letters of the wetland name if one word – if more than one word use the first letter of each word plus as many letters are needed in the last word (see 2.a).

(2) Add underscore_W.

(3) Use a 3-character stream mile including one whole number, the decimal and a tenth space. For example river mile 1.2 would be written as 1.2.

(4) Use the two letter county or state abbreviation from Appendix A. If the station is in another state, add an underscore _ before the two letter state abbreviation.

Example 1: A station located at DUCK wetland would be DUCK_W1.2CH.

Example 2: A station located at BLACK HORSE wetland would be BHORS_W1.2CH.

b. For unnamed wetlands with an associated stream

(1) Use the first five letter of the stream associated with the wetland if one word – if more than one word use the first letter of each word up to five letters (see 2. a.).

(2) Add underscore_W

(3) Use a 3-character stream mile including one whole number, the decimal and a tenth space. For example river mile 1.2 would be written as 1.2.

(4) Use the two letter county or state abbreviation from Appendix A. If the station is in another state, add an underscore _ before the two letter state abbreviation.

Example 1: A wetland associated with a stream Clear Creek would be CLEAR_W1.2SM.

c. For isolated unnamed wetlands with no stream associated with it, use the name associated with the ARAP permit request.

(1) Use the first five letters of the company associated with the wetland, - if more than one word use the first letter of each word up to five words.

(2) Add underscore_W.

(3) Use a 3-character stream mile including one whole number, the decimal and a tenth space. For example river mile 1.2 would be written 1.2.

(4) Use the two letter county or state abbreviation from Appendix A. If the station is in another state, add an underscore _ before the two letter state abbreviation.

Example: Company name Boones Farm BFARM_W1.2CO

5. Sinking streams

a. For named sinking streams

(1) Use the first five letters of the stream name if one word – if more than one word use the first letter of each word up to five words.

(2) Add underscore _S.

(3) Use a 3-character stream mile including one whole number, the decimal and a tenth space. For example, river mile 1.2 would be written as 1.2. The underground mileage between surface points will need to be estimated.

(4) Use the two letter county or state abbreviation from Appendix A. If the station is in another state, add an underscore _ before the two letter state abbreviation.

Example 1. A station located on Dry Creek would be DRY_S1.2CU.

Example 2. A station located on Stinky Cow Creek would be SCOW_S1.2CU.

Example 3. A station located at the sinkhole on Stinky Cow Creek would be SCOW_S0.0CU

b. For unnamed sinking streams

(1) Use the first five letters of the closest named geographical feature if one word – if more than one word use the first letter of each word up to five words.

(2) Add underscore _S.

(3) Use 1T for the first unnamed tributary that sinks into the sinkhole, 2T for the second unnamed tributary that sinks into the sinkhole, etc.

(4) Use a 3 character stream mile including one whole number, the decimal and a tenth space. For example, river mile 1.2 would be written as 1.2. Estimate underground mileage between surface points.

(5) Use the two letter county or state abbreviation from Appendix A. If the station is in another state, add an underscore _ before the two letter state abbreviation.

Example 1. An unnamed sinking stream station located on Crane Top Ridge would be CTOP_S1T1.2FR

6. Reservoirs (man-made lakes)

a. Assign the first 5 letters of the impounded stream (or embayment).

b. Use a 5 character stream mile if the sample is collected near the river channel. If the sample is collected near the right or left bank (such as at a boat dock) use a 4 character stream mile and the letter L or R to designate the right or left descending shore.

c. Use the appropriate 2 letter county or state abbreviation from Appendix A. Add an underscore _ before the two letter state abbreviation for stations in another state.
For example, a station that was collected from a boat on Fishing Lake which dams Otter Creek in Anderson County would be OTTER012.3AN. If the station was collected off a dock near the left descending shore the station ID would be OTTER12.3LAN.

In the site description include the reservoir name as well as location for clarification (for example Otter Lake near boat dock).

7. Natural Lakes

   a. Use the first 5 digits of the lake's name

   b. Using an S to designate station and a two digit whole number, assign the next available station number. For example if station numbers 1 through 4 already exist on that lake from previous studies (check the water quality database) then use station number 5. This would be designated S05.

   c. Use the appropriate 2 letter county or state abbreviation from Appendix A. Add an underscore _ before the two letter state abbreviation for stations in another state.

      For example, a new station located on Reelfoot Lake in Obion County would be REELFS11OB .

8. Duplicates

   A duplicate sample will be labeled with the appropriate station's ID and FD at the end.

   For example, if a duplicate sample was taken at Puncheoncamp Creek at river mile 1.5, the label would read PUNCH001.5GE-FD.

Protocol C –Buffered Formalin Preparation

Biologist/Environmental Specialist

1. Place a clean weigh pan on the balance. Tare the balance.

2. Weigh out approximately 1 gram of sodium bicarbonate for each liter of formalin.

3. Use extreme caution when handling formalin. It is a very toxic chemical and can cause irritation to skin, eyes, and respiratory system. It is also highly flammable. A lab coat, latex or vinyl gloves, and eye protection are required when working with formalin. Avoid breathing vapors or exposing co-workers to them. Work in a well ventilated area (preferably a lab hood or chemical ventilation system). Keep face away from open containers of formalin. Refer to the MSDS for information and first aid measures for reagents and chemicals in this SOP.

4. Gradually add the sodium bicarbonate to your container of formalin (37%). In general, one gram of sodium bicarbonate will buffer about one liter of formalin, but it is necessary to check the pH of the buffered solution, as individual batches of formalin may vary in pH. Close the container and shake vigorously after each addition. Check the pH at each interval until reaching a pH between 7.5-8.0 S.U. The sodium bicarbonate may not all dissolve into the formaldehyde, as this is a supersaturated solution. The solution should not be milky white; this indicates that too much sodium bicarbonate was added. Store the buffered formalin in an appropriate container such as Nalgene plastic and label the container "Buffered Formalin" and also write the date of preparation. The shelf life for the buffered formalin is two years.

Protocol D – Field Parameters

Biologist/Environmental Specialist

Adapted from U.S. Environmental Protection Agency. 2002

Measure dissolved oxygen, pH, temperature and conductivity at each periphyton monitoring station before flow is measured and macroinvertebrate or periphyton samples are collected. Allow readings to equilibrate before recording measurements. Record the duplicate readings in the appropriate area on the RPS sheet.

Label all meters as property of the State of Tennessee, Department of Environment and Conservation. Assign each meter a distinct identifying designation, (i.e. letter or a portion of the serial number) for calibration, maintenance, and deployment records. Mark each meter with this designation. Record the meter's ID number on the Rapid Periphyton Survey sheet (RPS) or Stream Survey Sheet. Multi-probe or individual meters meeting the following minimum specifications may be used (Table 9). Beyond following the instructions in this SOP for calibrating, maintenance, and logging procedures, it is also recommended to refer to manufacturer's instructions.

Table 2: Instantaneous Probe Minimum Specifications

Parameter	Range	Accuracy	Resolution
Temperature	-5 ^{o}C to 45 ^{o}C	+/- 0.20 ^{o}C	0.1 ^{o}C
Specific Conductivity	0 to 100,000*umhos/cm	+/- 1% of reading	4 digits
pH	2 to 12 units	+/- 0.2 units	0.01 units
Dissolved Oxygen	0 to 20 mg/L	+/- 0.2 mg/L	0.01 mg/L

* Areas of mining or other high conductivity/low pH may need a higher range.

1. **Calibrate Meter(s)** – Meters only need to be calibrated if they are going to be used that week. At the beginning of each week or day, or within 24 hours of use, in the EFO lab, calibrate meter(s) for all parameters that will be measured, following the manufacturer's instructions. Conductivity and pH probes are calibrated weekly with a drift check performed daily upon return (or at the end of the sampling period if overnight travel is involved). The drift check can be performed the next morning if time is a factor. The probes must be recalibrated when the drift check is out of the acceptable range, otherwise calibrating these probes once a week is acceptable. A drift check should be performed weekly for temperature. DO probes are to be calibrated each morning of use and at each site where necessary (see # 2). Drift checks for DO probes are not necessary if the meter was recalibrated in the field. If probes are factory calibrated, check readings against the appropriate standards to ensure the calibration is still accurate. Maintain calibration SOPs for each type and/or brand of meter. Keep all calibration records in a bound logbook (Figure 1). Include the date, meter designation, project name/number, initials of calibrator, parameter, standards used, meter reading, and adjustments. Also, record routine maintenance and repairs in the logbook. Some probes must be sent to the manufacturer for

calibration. Other probes must be replaced when they no longer maintain their calibration. In these cases, refer to manufacturer's instructions.

To check the calibration of the temperature probe place an ASTM thermometer in a container of room temperature water large enough to submerge the temperature probe. Place the meter in the water bath and allow it to equilibrate then compare the probe's reading to the thermometer's reading and mathematically adjust the probe's temperature as necessary. Coordinate with TDH laboratory to include the ASTM thermometer in their annual thermometer calibration check against the ASTM certified thermometer. Record this information in the calibration log.

EFO Meter Calibration Log

Date	Meter	Project	Init.	Parameter	Standard	Reading	Adj	Comments
3/6/02	YSI-A	Davis Ck	JEB	Conductivity	142	120	142	Cleaned contacts
3/6/02	YSI-A	Davis Ck	JEB	Conductivity	142	140	NA	Drift Check

Figure 1: Example of Meter Calibration Log

2. **Calibrate DO Probe** – The DO probe must be calibrated using either Winkler Titration (mg/l) or air calibration (% saturation) each morning prior to use. Most probes automatically compensate for temperature changes. Some probes also automatically compensate for pressure changes. An ASTM r calibrated thermometer and/or a handheld barometer must be carried in the field if the probe does not compensate for temperature and/or pressure changes. It is only necessary to recalibrate the probe at sites where there is a significant elevation, pressure or temperature change and the meter does not automatically compensate. A significant change in elevation is 1000 feet. A significant change in pressure is ±20 mm Hg (higher or lower) or when a storm front comes through the area. A significant change in temperature includes any ±5ºC change in temperature (higher or lower). If the DO probe is air calibrated, changes in pressure do affect concentration readings. Record the air calibration at the site in a calibration log in the field to the specified resolution in Table 9.

3. **Probe Placement** – Ideally, measure water parameters before measuring flow or collecting other samples (i.e. macroinvertebrate, periphyton). Turn on the meter(s) and if there is a DO stirrer, be sure it is activated. Carefully place the meter(s) in the thalweg upstream of the chemical and bacteriological sampling area. Suspend the probe(s) in the water column so it does not touch the bottom. If the water is too shallow to suspend the meter(s), carefully lay it on its side on firm substrate (preferably rock). Do not allow the probe(s) to sink into soft substrate.

Stand downstream of the probe, being careful not to disturb the substrate in the area of the probe(s). Allow enough time for each reading to stabilize before it is recorded. Depending on the meter, it may take a couple of minutes for dissolved oxygen to equilibrate. Record initial readings in the field notebook or the field survey form to the specified resolution (Table 9). The multi-parameter probe may also be placed in a bucket filled with surface water with the DO stirrer activated and allowed to equilibrate. Rinse the bucket and probe once with surface water before placing probe in the bucket of water.

4. **Duplicate Readings** – Take duplicate measurements at each site. If time is a constraint (short sample holding times or daylight), duplicate readings may be reduced to the first and last site each day. To take a duplicate measurement, lift the probe completely out of the water, wait for the readings to change, then return it to the original location or slightly upstream if the sediment was disturbed. Allow the meter to equilibrate before recording readings. If the readings are off by more than 0.2 units for pH, temperature, and DO in mg/L or off by more than 10% for specific conductivity, repeat the procedure until reproducible results are obtained. Record all readings on the RPS sheet and in the field notebook or the stream survey form. All results are to be recorded to the resolution specified in Table 9.

5. **Record Field Parameters** – Document the field parameters on the RPS form (Appendix B). Specific conductivity must be recorded in umhos/cm or uS/cm, dissolved oxygen in ppm (mg/l), and temperature in ^{o}C. If meter readings are in other units, record the exact readings on the RPS sheet and in the field survey form or field book and record the converted readings.

6. **Drift Check** – Without post-calibration checks, the accuracy of the water parameter measurements cannot be demonstrated. At the EFO lab, perform a drift check on each meter at the end of the day (or at the end of the trip on multiple night trips) and record results in the logbook (Figure 1). Drift checks can be done in the field as long as you have the proper equipment. To check the probes have maintained their calibration for pH and conductivity, compare the probe's readings against the appropriate pH, and conductivity standards. Adjust calibration if the probe is going to be used again that week. If the meter's calibration is off by more than 0.2 for pH or more than 10% for conductivity, all readings between the initial calibration and the drift check must be marked as questionable (N). To check the probes have maintained their calibration for temperature, compare the probe's readings against a standard ASTM thermometer. If the meter's calibration is off by more than 0.2, all the readings between the initial calibration and the drift check must be marked as questionable (N). When the DO probe has been air calibrated in the field due to pressure, elevation or temperature changes, a drift check is unnecessary at the end of the day. If the DO probe was not re-calibrated since leaving the base office, a drift check (Winkler or air calibration) should be performed at the end of the day. If the meter's calibration is off by more than 0.2 mg/L (Winkler) or 10% (air), all readings between the initial calibration and the drift check must be marked as questionable (N). On the Rapid periphyton Survey sheets (RPS), precede all questionable readings with an N (questionable data). If the RPS forms have already been submitted to TDH Environmental Laboratory, notify the Planning and

Standards Section in writing (e-mail or fax) of questionable readings so they may be noted in the Water Quality Database.

7. **Other Parameters** – some multi-parameter probes contain sensors for other water quality parameters such as turbidity or suspended solids. If these parameters are also measured, they should be calibrated following manufacturer's specifications prior to use with drift checks performed at the end of each day. Duplicate measurements should be taken at each site and recorded on the RPS sheet.

Protocol E – Habitat Assessment

Biologist/Environmental Specialist

Please note that a Standard Operating Procedure manual for Habitat Assessments will be published in 2010. When that document is published, it will supersede this protocol.

Conduct a habitat assessment any time a biological sample is collected. Use habitat data sheets finalized in *Rapid Bioassessment Protocols for Use in Wadeable Streams and Rivers* (Barbour et. al., 1999) to evaluate the integrity of the habitat at each site (Appendix B). When possible, two or more investigators should collaborate on habitat assessments to reduce the potential for individual bias.

Two different habitat assessment sheets will be used dependent on the ecoregion and/or stream type. (For habitat assessments, it does not matter if the upstream watershed is contained in the same bioregion). In order for the site to be compared to the habitat guidelines (Table 9), the assessment sheet used will depend on the ecoregion. Information on ecoregion boundaries can be found in the *Tennessee Ecoregion Project* (Arnwine et al, 2000). Each Environmental Field Office should have copies of ecoregion maps for their area. The Planning and Standards section should be contacted if there is uncertainty about what ecoregion a stream is located in.

In ecoregions 65j, 66d, 66e, 66f, 66g, 67f, 67g, 67h, 67i, 68a, 68b, 68c, 69d, 71e, 71f, 71g, 71h, and 74a as well as riffle streams in 71i, use the High Gradient Stream (formerly Riffle-Run) assessment sheet to evaluate habitat based on the guidelines in Table 9. Note that the guidelines cannot be used in non-riffle streams in these ecoregions. Therefore, a suitable upstream or watershed reference must be selected for comparison in non-riffle streams.

In ecoregions 65a, 65b, 65e, 65i, 73a, and 74b as well as non-riffle steams in 71i, use the Low Gradient (formerly Glide-Pool) assessment sheet. Copies of these sheets are located in Appendix B.

Evaluate all ten habitat parameters. Base score on a scale of 0 to 20 for each parameter, with 20 being the highest attainable score. Scores are divided into four categories (optimal, suboptimal, marginal and poor) with a range of five scores possible in each category. Specific guidance for scoring is located on the habitat sheets (Appendix B). The parameters that are evaluated in each sample reach are:

1. Epifaunal Substrate/Available Cover (high and low gradient streams)

Estimate the relative quantity and variety of natural structures in the stream such as cobble riffles, large rocks, fallen trees, logs and branches, and undercut banks that are available as refugia, feeding, spawning or nursery functions for macroinvertebrates and fish. Do not count “newly” fallen trees and unstable habitats.

2a. Embeddedness (high gradient streams)

Estimate the percent that rocks (gravel, cobble, and boulders) and snags are covered or sunken into the silt, sand, or mud of the stream bottom. To avoid confusion with sediment deposition (another habitat parameter), observations of embeddedness should be taken in the upstream and central portions of riffles and cobble substrate areas.

2b. Pool Substrate Characterization (Low gradient)

Evaluate the type and condition of the bottom substrate in the pools. Firmer sediment such as gravel and sand, and rooted aquatic plants support a wider variety of organisms and should be scored higher than a pool substrate dominated by mud or bedrock with no plants. In addition, a stream that has a uniform substrate will support fewer types of organisms and should score lower than a stream that has a variety of substrate types.

3a. Velocity/Depth Combinations (high gradient)

Determine the patterns of velocity and depth. The four basic patterns are slow-deep, slow-shallow, fast-deep, and fast-shallow. The best streams will have all four patterns present. The general guidelines are 0.5 meter depth to separate shallow from deep and 0.3 m/sec to separate fast from slow.

3b. Pool Variability (low gradient)

Rate the overall mixture of pool types found in the stream, according to size and depth. The four basic types of pools are large-shallow, large-deep, small-shallow, and small-deep. A stream having many different pool types will support a wider variety of aquatic species and should score higher. General guidelines are any pool dimension (length, width, oblique) greater than half the cross-section of the stream for separating large from small and 1 meter depth separating shallow and deep.

4. Sediment Deposition (high and low gradient)

Estimate the amount of sediment deposition. This is observable through the formation of islands, point bars (areas of increased deposition at the beginning of a meander that increase in size as the channel is diverted toward the outer bank) or shoals. Determine whether pools and runs are filling in. Usually deposition is evident in areas that are obstructed by natural or manmade debris and areas where the stream flow decreases, such as bends or pools.

5. Channel Flow Status (high and low gradient)

Estimate the degree to which the channel is filled with water. When water does not cover much of the streambed, the amount of suitable substrate for aquatic organisms is limited and the stream should score lower.

6. Channel Alteration (high and low gradient)

Determine how much, if at all, the stream has been altered. Channel alteration is present when artificial embankments, riprap, and other forms of artificial bank stabilization or structures are present; when the stream is very straight for significant distances; when dams and bridges are present; when dredging or gravel removal is evident and when other such artificial changes have occurred. Scouring is often associated with channel alteration.

7a. Frequency of Riffles or Bends (high gradient)

Determine the pattern of stream morphology by estimating the sequencing of riffles. In high gradient streams where distinct riffles are uncommon, a run/bend ratio can be used as a measure of meandering or sinuosity. In headwaters, riffles are usually continuous and the presence of cascades or boulders provides a form of sinuosity. To determine this parameter, a longer segment or reach than that designated for sampling should be incorporated into the evaluation.

7b. Channel Sinuosity (low gradient)

Evaluate the meandering or sinuosity of the stream. A high degree of sinuosity provides diverse habitat for macroinvertebrates and the stream is better able to handle surges when the flow fluctuates due to rain events. To estimate this parameter, a longer segment or reach than that designated for sampling should be incorporated into the evaluation. (This will vary by site, but should include at least two bends).

8. Bank Stability (high and low gradient)

Determine whether the stream banks are eroded or have the potential for erosion. Steep banks are more likely to collapse and suffer from erosion than are gently sloping banks, and are therefore considered less stable. Signs of erosion include crumbling, unvegetated banks, exposed tree roots, and exposed soil. Each bank is evaluated separately on a scale of 0 to 10 and the cumulative score of both banks is used for this parameter. (This parameter should be evaluated within the 100-meter sample reach.)

9. Bank Vegetative Protection (high and low gradient)

Determine the amount of vegetative protection afforded to the stream bank and near-stream portion of the riparian zone. The object is to determine the ability of the bank to resist erosion as well as the ability of the plants to uptake nutrients, control instream scouring, supply food to shredders and provide stream shading. Streams that have various types (shrubs, trees etc.) of native vegetation providing full natural plant growth will score highest. In some regions, the introduction of exotics, such as kudzu, has virtually replaced all native vegetation. The value of exotic vegetation to the quality of the habitat structure and contribution to the stream ecosystem should be evaluated, generally resulting in a lower score. Each bank is evaluated separately on a scale of 0 to 10 and the cumulative score of both banks is used for this parameter. (This parameter should be evaluated within the 100-meter sample reach.)

10. Riparian Vegetative Zone Width (high and low gradient)

Estimate the width of natural vegetation from the edge of the stream bank out through the riparian zone (approximately 18 meters). Disturbance to the riparian zone occurs when roads, parking lots, fields, lawns, bare soil, or buildings are near the stream bank. Residential developments, urban centers, golf courses, pastures and row crops are common causes of degradation of the riparian zone. However, the presence of old fields (previously grazed fields, not currently in use), paths, and walkways in an otherwise undisturbed riparian zone may be judged to be inconsequential to altering the riparian zone and may be given relatively high scores. Each bank is evaluated separately on a scale of 0 to 10 and the cumulative score of both banks is used for this parameter. (This parameter should be evaluated within the 100-meter sample reach.)

Total the 10 habitat parameters and compare the score to the Habitat Assessment Guidelines (Table 1) to determine whether the habitat is capable of supporting a healthy benthic community. Note that habitat scores in five ecoregions (68a, 68b, 68c, 69d and 71i) vary by season.

Sometimes it may be useful to evaluate individual parameters in addition to the total habitat score. For example even if the total habitat score meets regional guidelines, the individual parameters of embeddedness and sediment deposition may be low indicating a problem with sedimentation. Likewise, there may be a problem with riparian removal even though habitat scores meet regional guidelines. On the other hand, a low total score may not indicate a habitat problem if the channel flow status and velocity depth regime score low in a region where reference streams have extremely reduced flow during the summer and fall. Appendix A provides ecoregion specific expectations for each parameter on the Habitat guidelines form.

Table 3: Habitat Assessment Guidelines

Ecoregion	Habitat Form	Season	Not Impaired	Moderately Impaired	Severely Impaired
65a	Low Grad.	Jan. – Dec.	≥ 62	48 – 61	≤ 47
65b	Low Grad.	Jan. – Dec.	≥ 92	72 – 91	≤ 71
65e	Low Grad.	Jan. – Dec.	≥ 115	90 – 114	≤ 89
65i	Low Grad.	Jan. – Dec.	≥ 98	77 – 97	≤ 76
65j	High Grad.	Jan. – Dec.	≥ 158	124-157	≤ 123
66d	High Grad.	Jan. – Dec.	≥ 146	114 – 145	≤ 113
66e	High Grad.	Jan. – Dec.	≥ 143	113 – 142	≤ 112
66f	High Grad.	Jan. – Dec.	≥ 138	108 – 137	≤ 107
66g	High Grad.	Jan. – Dec.	≥ 173	130 - 172	≤ 129
67f	High Grad.	Jan. – Dec.	≥ 130	103 – 129	≤ 102
67g	High Grad.	Jan. – Dec.	≥ 117	92 – 116	≤ 91
67h	High Grad.	Jan. – Dec.	≥ 126	99 – 125	≤ 98
67i	High Grad.	Jan. – Dec.	≥ 120	95 - 119	≤ 94
68a	High Grad.	Jan. – June	≥ 156	130 - 155	≤ 130
68a	High Grad.	July – Dec.	≥ 139	103 – 138	≤ 102
68b	High Grad.	Jan. – June	≥ 144	113-143	≤ 112
68b	High Grad.	July – Dec.	≥ 109	86 - 108	≤ 86
68c	High Grad.	Jan. – June	≥ 128	101 – 127	≤ 100
68c	High Grad.	July – Dec.	≥ 121	95 – 120	≤ 94
69d	High Grad.	Jan. – June	≥ 160	126-159	≤ 125
69d	High Grad.	July – Dec.	≥ 164	129-163	≤ 128
71e	High Grad.	Jan. – Dec.	≥ 116	91 – 115	≤ 90
71f	High Grad.	Jan. – Dec.	≥ 123	97 – 122	≤ 96
71g	High Grad.	Jan. – Dec.	≥ 123	97 – 122	≤ 96
71h	High Grad.	Jan. – Dec.	≥ 117	92 – 116	≤ 91
71i	High/Low Grad.	Jan. – June	≥ 98	77 – 97	≤ 76
71i	High/Low Grad.	July – Dec.	≥ 96	76 – 95	≤ 75
73a	Low Grad.	Jan. – Dec.	≥ 94	74 – 93	≤ 73
74a	High Grad.	Jan. – Dec.	≥ 88	70 – 87	≤ 69
74b	Low Grad.	Jan. – Dec.	≥ 98	77 – 97	≤ 76

Protocol F – Rapid Periphyton Survey (RPS)

Biologist/Environmental Specialist

Every time periphyton sampling occurs, the sampler will need to complete a Rapid Periphyton Survey and a Multi-habitat Periphyton Sample (See Protocol G.) Completely fill out the front of the Rapid Periphyton Survey Data Sheet (Appendix B). Complete header information: Station ID, Sampler, and assign a log number. The back is not necessary if a Stream Survey Form (used for macroinvertebrates stream surveys) has been filled out, otherwise complete this form also. Add additional information not included on the sheet as needed. Only use the version provided in Appendix B. Earlier versions should no longer be used. Consult all personnel present during sampling for additional observations that may have been overlooked before leaving the site.

Front of RPS Data Sheet:

On the front of the Rapid Periphyton Survey Data Sheet is a form for recording algal biomass, canopy cover, and the habitats that were sampled for the MPS. This method has been adapted from EPA's Rapid Bioassessment Protocols for Use in Wadeable Streams and Rivers (1999).

1. Establish five transects across each stream reach. Target locations of likely algal growth (riffles and runs) if available. Avoid shaded areas if possible. Space transects to span full length of the reach.

2. Along each transect, observe 10 evenly spaced points. If the stream is not wide enough to observe 10 points (<1.5 m), observe as many points as possible and note the number of points observed and the reason in the comment field.

3. The Rapid Periphyton Survey Board is a 15 cm by 15 cm square of Plexiglas that has been divided into quadrants (see Figure 2). At each point on the transect, use the Rapid Periphyton Survey Board to estimate moss cover, macroalgal cover and microalgal biofilm thickness using the classification system defined in Table 4. The coverage class (0-5) for moss and macroalgae and the biofilm thickness (0-5) is to be recorded in the appropriate column beside the selected point on the transect on the RPS.

4. Optimal substrate is defined as substrate with a diameter greater than 2 cm. Smaller substrate is not as suitable for the growth of a stable population of algae due to scouring and shifting. At each point on the transect, determine if the substrate is of optimal size. If the substrate is not optimal in size for a stable periphyton population, but moss, macroalgae, and/or microalgae are visible, score and record that information on the RPS sheet in the comments.

5. At the mid-point of each transect, take a canopy reading using a spherical densiometer. The densiometer is a convex mirror etched into 24 ¼-inch boxes (Figure 3). Each box can be subdivided into 4 smaller squares, via an imaginary dot in the center of the box, to create a total of 96 smaller squares that can be counted within the entire densiometer.

Hold the densiometer 1 foot above the water surface. Holding the instrument at this level eliminates errors due to differing heights of samplers and different water depths, and includes low overhanging vegetation more consistently than holding the densiometer at waist level. Take four measurements, facing upstream, downstream, the right descending bank, and the left descending bank. Hold the instrument far enough away from the body so that the operator's head is just outside the grid. Count the number of small squares (out of a total of 96) that have tree canopy. Record this number (number of dots WITH canopy cover) on the datasheet. In order to get the overall percent canopy cover for that point, sum the four measurements and divide the total by 384.

6. Record the number of aliquots taken in each habitat type in the "Habitats Sampled" field.

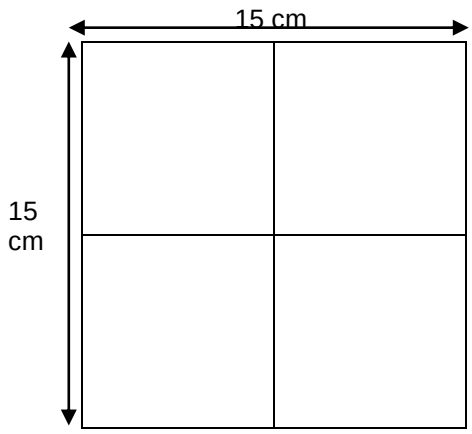


Figure 2: Rapid Periphyton Survey Board

A: 

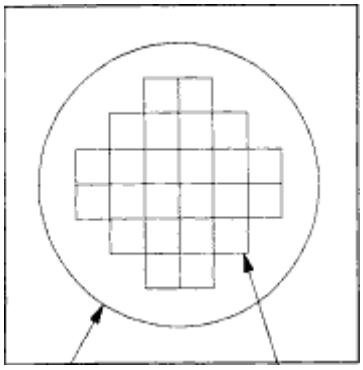
B: 

Figure 3: Spherical Densiometer
A: Picture of densiometer. Picture provided by Joellyn Brazile, Memphis EFO.
B: Grid etched onto the mirror. Picture from Cook et al, 1995.

Table 4. Percent Cover and Thickness Classes

Moss and Macroalgae Cover Classes						
Class Number	0	1	2	3	4	5
Coverage	0%	<5%	5% to 25%	26% to 50%	51% to 75%	>75%
Microalgal Thickness Class						
Class Number	0	1	2	3	4	5
Thickness	0 mm	<0.5 mm	0.5 to 1 mm	1 to 5 mm	5 to 20 mm	>20 mm
Characteristics	rough	Slimy, no visible biofilm	Biofilm visible			

Back of RPS Data Sheet (not required if Stream Survey Form has been completed):

1. Header Information – Complete all information in this section at new sites (those not already in the water quality database). Note that latitude and longitude is recorded in decimal format. Only the station number, stream name, station location, assessors, date and time need to be completed on existing sites already in the water quality database, or if a stream survey form is filled out because a macroinvertebrate sample is being collected as well. Record the time that the RPS was started in the header and the time the field measurements (D.O., pH, Conductivity, Temperature) were taken under Field Measurement. Use the 24-hour clock to designate time.

2. Field Measurements – Use calibrated meters for all field measurements. Designate what type of meter (and which meter) was used to make readings. The readings for each parameter (including duplicates) are recorded in the appropriate boxes. All readings are recorded in the units specified on the sheet. List any other readings, such as percent oxygen saturation, that were taken at the time of sampling (include units). Also, record field readings on the chemical request forms if chemical samples are being collected at the same time. (Average duplicate readings on the chemical request forms since only one value can be entered in the laboratory's system.) This ensures readings will be received by the Central Office and will be entered in the Water Quality Database.

If, after the drift check, the meter was found to be off by more than 0.2 units for pH, temperature, or dissolved oxygen (or more than 10% for conductivity), write an N before the reading on the field data sheet (RPS or Stream Survey Form) for all sites visited between the initial calibration and the drift check or discard data. The N designates questionable readings. Also, put an N before readings on the chemical request form. If the chemical request form has already been turned into the laboratory, fax the field data sheet to the Central Office. This will ensure the readings are flagged as questionable when they are entered into the Water Quality Database.

Circle the appropriate level of precipitation for the previous 48 hours (or circle unknown). Also, indicate the ambient weather.

3. Sediment Deposits – Circle the appropriate level of sediment deposits for the site. Also indicate the type of sediment found there, and the level of turbidity.

4. Stream Sketch– A station sketch is made at the time of sampling. This sketch should be detailed enough so that subsequent sampling teams or data reviewers can determine where samples were taken and what potential sources of impairment were present. Use a separate sheet of paper if necessary. At a minimum, the sketch should include a rough outline of stream sinuosity, direction of flow, location of riffles and pools, location of samples (benthic, periphyton, chemical, probe), location of bridges or any other man-made structures (include distance from sampling point), location of tributaries, run-off ditches, discharges, livestock access, and any other potential pollution sources. If a stream sketch has already been done on one of the other forms there is no need to duplicate, just indicate what form it is on.

5. Additional Comments – Describe any other conditions observed at the time of sampling (additional pages are to be used if necessary). Include any changes observed from previous sampling efforts. Note directions to the site and any special permission or keys needed for access. Ask other team members for input.

Protocol G – Multi-habitat Periphyton Sample (MPS)

Biologist or Environmental Specialist

Every time periphyton sampling occurs, the sampler will need to complete a Rapid Periphyton Survey (see Protocol F) and complete a Multi-habitat Periphyton Survey. There are two different sampling methods for the Multi-habitat Periphyton Survey that are used based on substrate type. One method is for non-sediment substrates and is the standard method of collection. The alternative method is for substrates that are sediment/sand and is only used if no other productive habitats are available. The number of aliquots collected using the standard method should be proportional to the substrate types found in the stream.

1. Examine the habitat for optimal periphyton collection.

   a. Standard Method

      - Identify target substrates where algae can be collected within arm's length (0.5 m) of the surface.
      - Typical substrates include removable portions of vascular plants or mosses, snags, roots, leaf packs, and rocks.
      - Substrate should be "seasoned" (not recent or "new fall").
      - Substrate should be exposed to sunlight.
      - Every effort should be made to collect from undisturbed areas. Avoid areas of recent walking or scouring.
      - Sampling should be conducted at low flow conditions, at least 1 week after high flow events.
      - Equally apportion substrates into 10 aliquots.
      - See Table 5 for examples of aliquot apportionments.

   b. Alternative Method - used for sampling in sediment (only if no other productive habitats are available)

      - Identify target substrates that are representative of different sediment conditions present along the entire reach of the stream.
      - Sediment condition factors to consider: Erosional, depositional, current velocity, and sunlight exposure.
      - Only consider sediment areas that are within arm's length (0.5m) of the surface.
      - Every effort should be made to collect from undisturbed areas. Avoid areas of recent walking or scouring.
      - Sampling should be conducted at low flow conditions, at least 1 week after high flow events.
      - Equally apportion substrates into 10 aliquots.

2. Collect Sample

a. Standard Method (Scrape)

- Fill the 500 mL wide mouth sample jar with site water to 100 mL fill mark.
- Carefully remove target substrate from water (must break or cut, so select smaller snags, removable roots, etc.) Do not limit rock size to sample jar diameter.
- Remove algae from substrate.
- Using fingers, rub substrate surface in the 100 mL of site water.
- The amount of surface area rubbed should be approximately 9 cm in diameter. Use mouth of 500 mL sample jar to estimate.
- Rub the entire substrate surface a minimum of three times to ensure algae removal.
- Rinse fingers in the jar.
- Discard substrate.
- Take 2 sub-samples and composite into the 125 mL amber sample bottle.
  - Stir well to homogenize periphyton in the 100 mL slurry.
  - Remove first 2 mL subsample with a disposable pipette and transfer into sample bottle. The pipette needs to be completely filled.
  - Repeat stirring procedure in 100 mL slurry.
  - Remove second 2 mL subsample with pipette and transfer into sample bottle.

b. Alternative Method (Syringe)

- At the selected sample point, pull the plunger of the syringe core back to the 3 cm mark (15 mL) and insert the open end of the syringe core into the surface of the sediment until the core is level with the sediment surface.
- Try to be extremely delicate, so that the overlying sediment will be disturbed as little as possible.
- Slide the spatula through the sediment to cover the opening of the syringe core.
- Remove the syringe core and spatula together, and invert positioning so that the spatula is on top of the syringe core opening.
- Using the plunger, push sample into the 500 mL wide-mouth sample jar.
- Repeat the previous steps until all of the sediment samples are collected and contained in the 500 mL wide-mouth sample jar.
- Use the measuring spoon (tablespoon) to homogenize the sample in the beaker until the sediment is consistent in color and texture throughout.
- Using the tablespoon, remove 1 flat spoonful of sediment and place into the 125 mL opaque amber plastic wide-mouth sample bottle.
- Place enough water in the sample bottle so that the sediment is submerged.

3. Add 2 mL of buffered formalin (37%, see Protocol C for preparation) to the periphyton sample bottle.

4. Label sample with identifying information.

- Complete an external tag (Standard external tag provided by the state lab) and attach to the outside of the container (Figure 4).
- Write the Sample Log number (see Protocol H) in the "Field #" space on the external tag.
- Be sure to record the number of aliquots taken in each substrate type on both the RPS (in the "Habitat Sampled" field) and the sample tag (in the "Remarks" field).

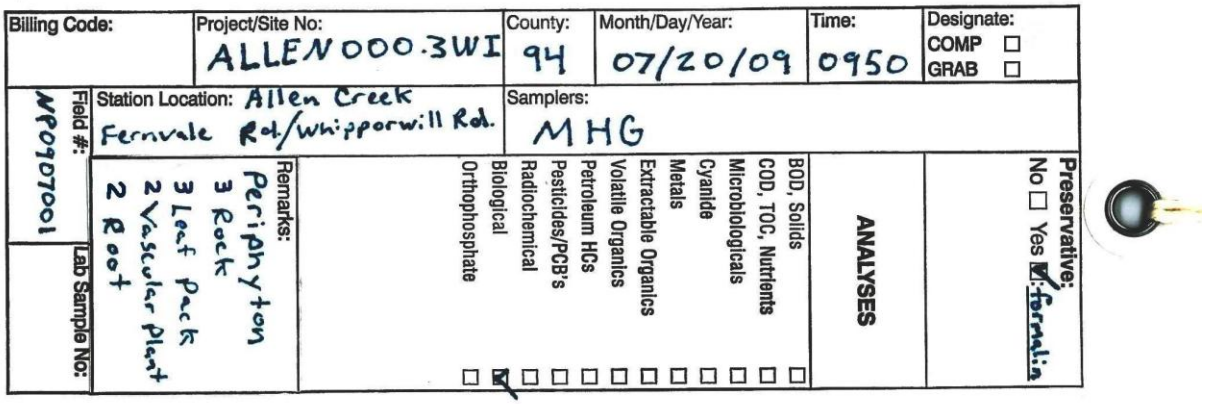


Figure 4: Example of External Tag

Table 5. Example of Aliquot Apportion Strategies

Number of Productive Substrates	Aliquot Apportion Strategy
1	10 aliquots from single substrate
2	5 aliquots from each substrate
3	4 aliquots from most abundant substrate 3 aliquots from remaining two substrates
4	3 aliquots from two most abundant substrates 2 aliquots from remaining two substrates
5	2 aliquots from each substrate
6	2 aliquots from four most abundant substrates 1 aliquot from two remaining substrates

Protocol H: Sample Logging

Any staff member

Samples must be logged to allow complete reconstruction, from initial field records, through data storage and system retrieval. The sampler must assign a discrete log number to each individual periphyton sample. This will be a nine digit number determined in the following manner:

NP0105001

The first digit (N) determines which office the sample is from:

C = Chattanooga EFO	K = Knoxville EFO
L = Columbia EFO	M = Memphis EFO
V = Cookeville EFO	N = Nashville EFO
H = Johnson City EFO	S = Surface Mining
J = Jackson EFO	B = Lab Services, TDH

The second digit will be a P to indicate this is a periphyton sample.

The third and fourth digit represent the year sampled (02 = 2002)

The fifth and sixth digit represent the month sampled (03 = March)

The last three digits represent a consecutive number for the number of samples collected that month (001 = the first sample collected in March).

The log numbers (along with the station number) will be used to identify the sample on paperwork, tags, bench sheets, logbooks, QC records, or any other place this sample is documented.

All samples are logged in a bound logbook. Make entries in black or blue ballpoint pen. (Note: a computer can be used to log Field Office samples; however, a hard copy printout in a bound logbook should also be maintained.) The log entry must include the sample log number, station ID, date collected, time collected, collector's initials, sample source, station location, type of sample and date sent to lab. The Lab log must include the sorters initials, sorting date, taxonomist's initials and ID date (figure 5).

Sample Log #	Station ID	Source	Location	Date Col.	Time Col.	Init. Col.	Date sent to Lab	Sorters Initials	Sorting Date	Taxonomist Initials	ID Date
NP0907001	ALLEN000.3WI	Allen Creek	Fernvale Rd/Whipporwill Rd	7/20/09	0950	MHG	07/21/09				
KP0203002	DAVIS001.3CL	Davis Ck	Hwy Z	3/6/02	1500	JEB/DRM	3/7/02				

Figure 5: Example of Periphyton Sample Log

Protocol I - Taxonomy of Periphyton Samples

Laboratory biologist or contractor with demonstrated expertise in periphyton taxonomy

All Periphyton samples are to be sent to the central lab for analysis. This is to be coordinated through the Planning and Standards Section.

1. The concentrated preserved periphyton sample will be divided for analysis. A 10mL subsample will be analyzed for non-diatom algae, and a separate 10 mL subsample will be analyzed for diatoms.

a. Non-diatom ("Soft") Algae Relative Abundance and Taxa Richness

1. Homogenize the subsample with a tissue homogenizer or blender.

2. Thoroughly mix the homogenized subsample and pipette into a calibrated (known volume) counting chamber for quantitative counting of cells, such as a Nannoplankton chamber or a Palmer-Maloney Counting Cell or other.

3. Fill in the top portion of the Non-diatom Algae Taxonomic Bench Sheet for "soft" algae (Appendix B).

4. Identify and count 300 algal "cell units" to the lowest possible taxonomic level at 400x magnification.
    - "Cell units" for coenocytic algae or filamentous algae can be defined as 10 µm sections of the thallus or filament.
    - In the non-diatom subsample, count only live diatoms. Identify these only to phylum; do not identify diatoms to a lower taxonomic level.
    - Record numbers of cells or cell units observed for each taxon on the bench sheet
    - Make taxonomic notes and drawings on bench sheets of important specimens.

5. Relative abundances of "soft" algae are determined by dividing the number of cells (cell units) counted for each taxon by the number of cells counted (e.g., 300). Enter this information on the Non-diatom Algae Taxonomic Bench Sheet (Appendix B).

6. Measure the linear dimensions of the first 25 cell units of each taxon found. To calculate biovolume, use the median of those 25 linear dimensions. If fewer than 25 cell units are found for that taxon, then measure all cell units.

7. Use the tables in Appendix E to calculate biovolume for each taxon. Enter this information on the Non-diatom Algae Taxonomic Bench Sheet (Appendix B).

b. Diatom Relative Abundance and Taxa Richness

1. Under a fume hood, add enough concentrated nitric or sulfuric acid to the subsample to produce a strong exothermic reaction. Usually equal parts of sample and acid will produce such a reaction. Caution: Use a fume hood, safety glasses, and protective clothing. Separate the sample beakers by a few inches to prevent cross-contamination of sample in the event of overflow.

2. Allow the sample to oxidize overnight.

3. Fill the beaker with distilled water.

4. Wait 1 hour for each centimeter of water depth in the beaker.

5. Siphon off the supernatant and refill the beaker with distilled water. Siphon from the center of the water column to avoid siphoning light algae that have adsorbed onto the sides of the water column.

6. Repeat steps 4 through 6 until all color is removed and the sample becomes clear or has a circumneutral pH.

7. Mount diatoms in Naphrax or another high refractive index medium to make permanent slides. Label slides with the same information as on the sample container label.

8. Fill in the top portion of the Diatom Algae Taxonomic Bench Sheet (Appendix B).

9. Identify and count diatom valves to the lowest possible taxonomic level, which should be species and perhaps variety level, under oil immersion at 1000x magnification. Count 600 valves (300 cells). Be careful to distinguish and count both valves of intact frustules. Record numbers of valves observed for each taxon on the bench sheet. Make taxonomic notes and drawings on the Diatom Algae Taxonomic Bench Sheet (Appendix B) of important specimens.

10. Determine the relative abundances of diatom species in the algal assemblage by dividing the number of valves counted for each species by the total number of valves counted (e.g., 600); then multiply the relative abundance of each diatom taxon in the diatom count by the relative abundance of live diatoms in the count of all algae. Enter this information on the Diatom Algae Taxonomic Bench Sheet (Appendix B).

11. Measure the linear dimensions of the first 25 cell units of each taxon found. To calculate biovolume, use the median of those 25 linear dimensions. If fewer than 25 cell units are found for that taxon, then measure all cell units.

12. Use the tables in Appendix E to calculate biovolume for each taxon. Enter this information on the Diatom Algae Taxonomic Bench Sheet (Appendix B).

Protocol J – Data Reduction of Periphyton Samples

Biologist/Environmental Specialist

Diatom Bioassessment Index

Because this is a new program for Tennessee, a periphyton bioassessment index has not been developed yet. It will take several years to accrue enough data points to develop a comprehensive index. Until that time, Kentucky's Diatom Bioassessment Index (KDBI) will be used for assessing diatom populations (KDEP, 2008).

The KDBI is similar to the TMI (Tennessee Macroinvertebrate Index) in that it is a multimetric index. It is based on the six individual metrics listed below:

1. Total Number Diatom Taxa (TNDT)
This metric measures diatom species richness. It is expected to decrease with increasing pollution.

2. Shannon Diversity (H')
The mean Shannon diversity index was chosen primarily because it is commonly used by aquatic biologists, so values will be more readily interpreted and compared with other literature values. Using this index, H′= 0 when only one species is present in the collection, and H′ is at a maximum when all individuals are evenly distributed among all species.

$$H' = -\sum \frac{n_i}{N} \log_{10} \frac{n_i}{N}$$

where:
n_i = number of individuals of species i
N = total number of individuals

3. Kentucky Pollution Tolerance Index (KPTI)
Kentucky's Pollution Tolerance Index (KPTI) is a measure of the overall tolerance level of the entire diatom community. A healthy population will include diatoms at all tolerance levels; however, the number of tolerant organisms should be comparatively low. The KPTI measures both the tolerance level of individual taxa and the overall abundance of those taxa. Tolerance values are included in Appendix D.

$$KPTI = \sum \frac{n_i t_i}{N}$$

Where:
n_i = number of individuals of species i
t_i = tolerance value of species i
N = total number of individuals in sample

4. *Cymbella* Group Richness (CGR)
This metric measures the total number of taxa represented in the sample from the genera *Cymbella, Cymbopleura, Encyonema, Encyonopsis, Navicella, Pseudoencyonema* and *Reimeria*. As water pollution increases, the CGR is expected to decrease.

CGR=*Cymbella + Cymbopleura + Encyonema + Encyonopsis + Navicella + Pseudoencyonema + Reimeria*

5. *Fragilaria* Group Richness (FGR)
This metric measures the total number of taxa represented in the sample from the genera *Ctenophora, Fragilaria, Fragilariforma, Pseudostaurosira, Punctastriata, Stauroforma, Staurosira, Staurosirella, Tabularia,* and *Synedra* reflect high water quality. As water pollution increases, the FGR is expected to decrease.

FGR=*Ctenophora + Fragilaria + Fragilariforma + Pseudostaurosira + Punctastriata + Stauroforma + Staurosira + Staurosirella + Synedra + Tabularia*

6. % *Navicula, Nitzschia* and *Surirella* (%NNS)
The sum of the relative abundances of all *Navicula* (including *Aneumastus, Cavinula, Chamaepinnularia, Cosmioneis, Craticula, Diadesmis, Fallacia, Fistulifera, Geissleria, Hippodonta, Kobayasia, Luticola, Lyrella, Mayamaia, Muellaria, Placoneis* and *Sellaphora*), *Nitzschia* (including *Psammodictyon* and *Tryblionella*) and *Surirella* taxa reflects the degree of sedimentation at a reach. These three genera are motile, using their raphes to slide through sediment if they become covered. Their abundance expresses the frequency and severity of sedimentation. As sedimentation increases, the %NNS is expected to increase.

$$\%NNS = \frac{\text{Number of } \textit{Navicula} + \textit{Nitzschia} + \textit{Surirella} \text{ taxa}}{\text{Total number of individuals in sample}} \times 100$$

Each metric is given a calculated score (range 0-100) based on the percent of the standard metric value (the 95th percentile for TNDT, H', KPTI, FGR, and CGR or 5th percentile for %NNS) of Kentucky's entire database (impaired and reference). These percentile thresholds are used to eliminate outliers. The formulae for calculating KDBI scores are shown in Table 6.

Table 6: Metric scoring formulae for the Kentucky Diatom Bioassessment Index

Metric	Formula
TNDT	(TNDT/102) X 100
H′	(H′/1.43) X 100
KPTI	(PTI/3.46) X 100
CGR	(CGR/13) X 100
FGR	(FGR/8) X 100
%NNS	(100 - %NNS)/(100 – 2.6) X 100

Metric scores with values greater than 100 receive a score of 100.0. The mean of the six KDBI metrics is the final KDBI score on a 0-100 scale.

The KDBI will be scored according to Table 7. For the purposes of assessment, the state will be divided into 3 bioregions: Mountains (**MT**), Mississippi Valley-Interior Region (**MVIR**), and Pennyroyal (**PN**). The MT region is comprised of ecoregions 68a, 68b, 68c, 68d, 69d, 69e, 66d, 66e, 66f, 66g, 66i, 66f, 66k, 67f, 67g, 67h, and 67i. The MVIR region is comprised of ecoregions 73a, 73b, 74a, 74b, 65a, 65b, 65e, and 65i. The PN region includes ecoregions 71e, 71f, 71g, 71h, 71i, and 65j. These assessments are based on Kentucky's Bioassessment protocols, as mentioned previously. There is some question about the dependability of this index for some ecoregions. Kentucky does not have ecoregions 65a, b, e, or i, 66d, e, f, g, i, j, or k or 67f, g, h, or i. Therefore, those ecoregions have been grouped with the bioregion to which they are most similar. These metrics will be evaluated for Tennessee's ecoregions as soon as possible.

Table 7. Bioassessment guidelines for KDBI scores.

	MT	MVIR	PN
Excellent	75-100	57-100	67-100
Good	62-74.99	48-56.99	55-66.99
Fair	51-61.99	42-47.99	50-54.99
Poor	0-50.99	0-41.99	0-49.99

Other metrics will be tested in order to evaluate their usefulness in developing a biotic index for Tennessee's streams. A list of these metrics can be found in Appendix C.

Non-diatom ("Soft") Algae Metrics

At this time there is no index of biotic integrity developed for "soft", or non-diatom algae. A variety of metrics will be tested to develop an index for future use. A list of these metrics can be found in Appendix C.

Rapid Periphyton Survey Metrics

At this time, an index of biotic integrity has not been developed for Rapid Periphyton Surveys. A variety of metrics will be tested to develop an index for future use. A list of these metrics can be found in Appendix C.

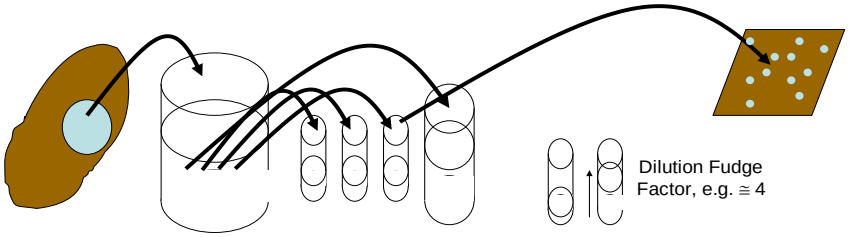
Area-Specific Cell Densities and Biovolumes

Cell densities are determined by dividing the numbers of cell counted by the proportion of sample counted and the area from which samples were collected. The **dilution fudge factor** is calculated by dividing the volume of each aliquot (4mL) by the volume of the water in which the substrate is scrubbed (100mL). The dilution fudge factor is 4% (or a value of 0.04 when used in the formula below. When the alternative MPS method is used, the cell density will be based on the volume of sediment collected instead of the surface area scrapped.

Cell biovolumes are determined by summing the products of cell density and biovolume of each species counted and dividing that sum by the proportion of sample counted and the area from which samples were collected. The formulas for biovolume calculations for each taxon (based on shape) can be found in Appendix E.

Calculating Cell Density

$$\frac{\text{cells counted}}{\text{sample area (or volume)} \times \frac{\text{vol. sample on c-glass}}{\text{total sample volume}} \times \text{Dilution Fudge Factor} \times \frac{\text{area c-glass counted}}{\text{area coverglass}}}$$



Spreadsheet Calculation Formula:

$$\frac{\text{cells counted}}{\text{sample area (or volume)}} \times \frac{\text{area coverglass}}{\text{area c-glass counted}} \times \frac{\text{total sample volume}}{\text{vol. sample on c-glass}} \times \text{DFF}$$

Protocol K – Report Preparation

Upon completion of a periphyton survey, the EFO will submit a copy of the Rapid Periphyton Survey data sheet, habitat assessment, map, and photos to PAS (also include stream survey form if completed). The EFO will submit the Sample Request Form, including the Chain of Custody to the lab along with the sample.

Once received at the Lab, a separate lab number will be assigned. Upon completion of sample, the lab will enter taxa into the SQSH database. The lab will submit a copy of the diatom taxa list, soft algae taxa list, sample request form, and periphyton assessment report to PAS and appropriate EFO.

I.J - DATA AND RECORDS MANAGEMENT

Results from periphyton samples (including taxa lists) are entered by the Aquatic Biology Section, Lab Services, TDH in an Access 2000 file called SQDATA (date). This database will be sent by the Aquatic Biology Section to the Planning and Standards Section at the completion of each project and/or quarterly.

Following processing and quality control checks, periphyton results are entered into Tennessee's Water Quality Database (WQDB), an Access file maintained by PAS. A copy of the WQDB is sent quarterly to EFO and CO personnel.

Assessment information for each stream segment will be entered in the Access 2000 Assessment Database (ADB) by the Planning and Standards Section (PAS). PAS staff will meet with WPC managers and biologists in each EFO before assessments are finalized. This database will eventually be accessible on the web for public access.

The original field sheets and taxa lists are to be kept in files at the sampling agency (Environmental Field Office or Aquatic Biology Section). Copies of the Non-Diatom Algae Taxonomic Bench Sheets (Appendix B), Diatom Algae Taxonomic Bench Sheets (Appendix B), habitat assessment sheets (Appendix B), Rapid Periphyton Survey Data Sheets (Appendix B), Periphyton Assessment Report (Appendix B), map of sample location, Chain of Custody forms (Appendix B), and photographs are to be sent (printed in color, emailed, or on CD) to the Planning and Standards Section. These copies will be kept in the watershed files for five years before being archived.

II. QUALITY ASSURANCE / QUALITY CONTROL

The U.S. EPA requires that a centrally planned, directed and coordinated quality assurance and quality control program be applied to efforts supported by them through grants, contracts or other formalized agreements. This time allocation is an essential component of biological sampling and analysis and will be included in annual work plans. This is not an optional or "as time allows" activity. The goal is to demonstrate the accuracy and precision of the biologists, as well as the reproducibility of the methodology, and to ensure unbiased treatment of all samples.

A. General QC Practices

1. Quality Team Leader (QC Coordinator) - A centralized biological QC coordinator will be designated with the responsibility to ensure that all QC protocols are met. This person will be an experienced water quality biologist. Major responsibilities will include monitoring QC activities to determine conformance, distributing quality related information, training personnel on QC requirements and procedures, reviewing QA/QC plans for completeness, noting inconsistencies, and signing off on the QA plan and reports.

2. Quality Team Member (In-house QC officer) - One WPC biologist/environmental specialist in each EFO will be designated as the Quality Team Member (in-house QC officer.) This person will be responsible for performing and/or ensuring that quality control is maintained and for coordinating activities with the central Quality Team Leader (QC coordinator).

3. Training - Unless prohibited by budgetary travel restrictions, training will be conducted at least once a year through workshops, seminars and/or field demonstrations in an effort to maintain consistency, repeatability and precision between biologists/environmental specialists conducting periphyton surveys. This will also be an opportunity for personnel to discuss problems they have encountered with the methodologies and to suggest SOP revisions prior to the annual SOP review. Note: topics of discussion should be submitted to the central Quality Team Leader (QC coordinator) before the meeting so that a planned agenda can be followed, thus making the best use of limited time.

B. Field Quality Control – Habitat Assessment and Biological Sampling Methodology

1. Habitat Assessments - At minimally 10% of sites, two trained biologists/environmental specialists will complete habitat assessment forms independently. Scores are compared for each parameter with discrepancies arbitrated while in the field.

2. Rapid Periphyton Survey - A second survey will be collected at a minimum of 10% of the sites by a separate biologist/environmental specialist. This should be conducted at the same time, or at least within two weeks of the original survey if flow characteristics are the same.

3. Periphyton Sample - A second multi-habitat periphyton sample will be collected at 10% of the sites.

4. Chain of Custody Chain of custody is required by the TDEC Office of General Counsel for samples that have the potential of being used in court, reviewed by state boards, or involved in state hearings. Chain of custody must also accompany any contract samples (periphyton samples being sent to the lab). Chain of custody is the far right column of the biological analysis form (Appendix B). The entire form must be filled out completely.

   The chain of custody follows the sample through collection, transfer, storage, taxonomic identification, quality assurance and disposal. The biologist who collected the sample must sign (not print) their name in full (not initial) in the Collected By space with the date and time (24-hour clock). If the sample is given to anyone else before it is delivered to the lab (or returned to the office), each person responsible for the sample must sign their full name on the Received By space with the date and time. The person in the laboratory (or office) who receives the sample will sign line four. The person who logs the sample in signs the last line.

C. QC Log

A list of all samples sorted and/or identified by each biologist/environmental specialist will be kept in a bound log or a backed-up computer spreadsheet/database so that QC requirements and results can be documented (Table 8). The QC log must contain the following information:

1. Sample log number
2. Station ID
3. Sample type
4. Initials of sampler and taxonomist
5. Date completed
6. Initials of person performing QC
7. Percent community similarity
8. Date of QC
9. Initials of QC taxonomist
10. Results of QC (satisfactory/unsatisfactory)

Table 8: Example of Periphyton QC Log

Sample Log #	Station ID	Sample Type	Sampler/ Taxonomist	Sample Date	QC'd By	QC Date	% Community Similarity	S/U
JP0201001	BIFFL003.0DY							
JP0201002	BIGGS000.7WY	MPS	AJF / KMM	3/11/02	PDS	3/20/02	95%	S
JP0201003	BMHOL002.0OB							
JP0201004	CANE001.8WY							
JP0201005	CGROU001.2WY							
JP0201006	CLEAR001.2HN							
JP0201007	CLOVE001.4OB							
JP0201008	CSPRI002.4DY							
JP0202001	CYPRE00.6WY							
JP0202002	CYPRE000.6OB							
JP0202003	DAVID002.6OB							
JP0202004	GRASS000.8OB							
JP0203001	HFORK006.8OB							
JP0203002	HOOSI000.5OB							
JP0203003	HURRI002.6WY							
JP0203004	HURRI003.9WY							
JP0203005	HURRI1T1.1WY							
JP0203006	MILL004.0OB							
JP0203007	NFOBI005.9OB	MPS	AJF/KMM	3/15/02	PDS	3/20/02	94%	S
JP0203008	NFOBI018.0WY							
JP0203009	NFOBI026.5WY							
JP0203010	NFOBI040.6HN		NA	NA	NA	NA	NA	NA
JP0203011	OBION020.9DY							
JP0203012	OBION044.3DY							

D. Percent Community Similarity

1. Each biologist/ES responsible for taxonomic identification, regardless of previous experience, will have every sample QC'd by a second biologist/ES who has already achieved 75% community similarity (documented) until the original biologist/ES has passed 75% community similarity on a sample. A record of this is kept in the QC log. Once a biologist/ES has passed their first QC, they are QC'd on 10% of subsequent samples (randomly selected). A community similarity of 75% must be maintained. If the community similarity is lower than 75%, every sample prior to that one in the same group of 10 is re-sorted until a sample that has met the 75% requirement is found. The next group of 10 starts after the unsatisfactory sample.

   The community similarity is calculated by:

   $$\text{Percent Community Similarity} = 100 - .5\sum_{i=1}^{s} \left| a_i - b_i \right| = \sum_{i=1}^{s} \min(a_i, b_i)$$

2. Log results in the QC log.

3. All QC must be completed before the data are released to ensure accuracy of results. If, for any reason, a report is released prior to QC completion, an addendum will be sent to all report recipients with any corrected information after QC is complete.

E. Reference Collections

1. A master collection of images of all taxa identified in the state will be kept in the central laboratory. The organisms in the centralized master reference collection will be verified by outside experts recognized for expertise in a particular taxonomic group. A list of verified organisms found in the state is provided in Appendix D. New taxa will be verified by an outside expert (Appendix D).

2. The central laboratory reference collection will be catalogued with discrete collection numbers assigned to each taxon. Assign sequential numbers to specimens as they are added into the collection. For example, 0001 would be the first specimen in the collection. Maintain an accession catalog of all reference material in a permanently bound log and on disk. Each entry must contain the following information:

   Accession number (This must be unique for each group of organisms in each collection)
   Complete name (genus, species, variety (if applicable), authority, date)
   Higher taxa (family, order, class)
   Locality data (Waterbody, site, county, ecoregion, station number)
   Sample type
   Name of collector/date of collection
   Name of taxonomist
   Name of verifier if appropriate
   Number of specimens

F. Data Reduction QC

1. Store raw data (non-manipulated) in one or more separate locations and in an electronic database with backup.

2. A second staff member checks all computer data entry correctness by direct comparison with the field or laboratory handwritten data sheets. The person performing the data entry QC initial and dates each page of the checked printout in red ink.

3. Keep QC information in a file with the other information for that project a minimum of five years.

4. Complete all data reduction QC before results are released.

III. REFERENCES

Bahls, Lauren L. 1993. Periphyton Bioassessment Methods for Montana Streams. Montana Department of Health and Environmental Sciences. Water Quality Division. Helena, Montana.

Barbour, M.T., Gerritsen, J., Snyder B.D. and J.B. Stribling. 1999. *Rapid Bioassessment Protocols for Use in Streams and Wadeable Rivers*. EPA 841-B-99-002. U.S. Environmental Protection Agency, Office of Water, Washington D.C.

DeLorme. 2004. *Tennessee Atlas & Gazetteer*. DeLorme, Yarmouth, Maine.

Fore, Leska S. and Cynthia Grafe. 2002. Using Diatoms to Assess Biological Condition of Large Rivers in Idaho (U.S.A.). *Freshwater Biology.* 47, 2015-2037.

Griffith, G.E., J.M. Omernik and S. Azevedo. 1997. *Ecoregions of Tennessee*. EPA/600/R-97/022. NHREEL, Western Ecological Division, U.S. Environmental Protection Agency, Corvalis, Oregon.

Hill, B.H., A. T. Herlihy, P.R. Kaufmann, R. J. Stevenson, F. H. McCormick, and C. Burch Johnson. 2000. Use of Periphyton Assemblage Data as an Index of Biotic Integrity. *Journal of the North American Benthological Society*. 19(1): 50-67.

Hillebrand, H., C. Durselen, D. Kirschtel, U. Pollingher, and T. Zohary. 1999. Biovolume Calculation for Pelagic and Benthic Microalgae. *Journal of Phycology*. 35: 403-424.

Kentucky Department of Environmental Protection. 2008. Standard Methods for Assessing Biological Integrity of Surface Waters in Kentucky. Division of Water. Frankfort, Kentucky.

U.S. Environmental Protection Agency. 1995. *Generic Quality Assurance Project Plan Guidance for Programs Using Community Level Biological Assessment in Wadeable Streams and Rivers.* EPA 841-B-95-004. U.S. EPA, Washington D.C.

Wang, Yi-Kuang, R. Jan Stevenson, Lythia Metzmeier. 2005. Development and evaluation of a diatom-based index of biotic Integrity for the Interior Plateau Ecoregion, USA. *Journal of the North American Benthological Society.* 24(4): 990-1008.

APPENDIX A

ECOREGION REFERENCE INFORMATION

ECOREGION REFERENCE STREAMS
REGIONAL EXPECTATIONS FOR INDIVIDUAL HABITAT PARAMETERS

ECOREGION REFERENCE STREAMS

SITE #	STATUS	STREAM	USGS HUC	LOCATION	COUNTY	LATITUDE	LONGITUDE
ECO65A01	Active	Unnamed Trib. to Muddy Creek	08010207 Upper Hatchie	RM 0.7 U/S Matt Dammonds Rd	McNairy	35.09583	-88.49944
ECO65A03	Active	Wardlow Creek	06040001 TN Western Valley	RM 4.6 Hamburg Rd	McNairy	35.02277	-88.44194
ECO65B04	Active	Cypress Creek	08010207 Upper Hatchie	RM 5.5 U/S Buster King Rd	Hardeman	35.0675	-88.86
ECO65E04	Active	Blunt Creek	06040005 TN Western Valley	RM 0.1 U/S McHee Levee Rd	Carroll	35.95916	-88.26805
ECO65E06	Active	Griffin Creek	08010204 S Fk Forked Deer	RM 5.1 U/S Stanford Lane Ford	Carroll	35.81861	-88.54055
ECO65E08	Active	Harris Creek	08010201 N Fk Forked Deer	RM 2.2 Potts Chapel Rd	Madison	35.62638	-88.69972
ECO65E10	Active	Marshall Creek	08010208 Lower Hatchie	RM 2.2 Van Buren Rd	Hardeman	35.1619	-89.0694
ECO65E11	Active	West Fork Spring Creek	08010208 Lower Hatchie	RM 1.7 U/S Van Buren Rd	Hardeman	35.10194	-89.08194
ECO65I02	Active	Battles Branch	06030005 TN Pickwick Lake	RM 0.8 U/S Old Kendrix Rd	Hardin	35.03333	-88.29305
ECO65J04	Active	Pompeys Branch	06030005 TN Pickwick Lake	RM 0.8 U/S Pompeys Branch Rd	Hardin	35.05388	-88.16805
ECO65J05	Active	Dry Creek	06030005 TN Pickwick Lake	RM 3.2 Dry Creek Rd	Hardin	35.035	-88.15222
ECO65J06	Active	Right Fork Whites Creek	06040001 TN Western Valley	RM 3.4 U/S Morris Lane	Hardin	35.05305	-88.04777
ECO65J11	Active	Unnamed Trib. Rt Fork Whites Cr	06040001 TN Western Valley	RM 0.1 U/S Morris Lane	Hardin	35.05225	-88.04825
ECO66D03	Active	Laurel Fork	06010103 Watauga	RM 6.7 U/S Big Branch Off Dennis Cove Rd	Carter	36.25694	-82.11111
ECO66D05	Active	Doe River	06010103 Watauga	RM 25.4 U/S Picnic Area Roan Mtn State Park	Carter	36.15888	-82.10583
ECO66E04	Active	Gentry Creek	06010102 South Fork Holston	RM 2.1 Gentry Creek Rds end.	Johnson	36.54444	-81.72444

ECOREGION REFERENCE STREAMS

SITE #	STATUS	STREAM	USGS HUC	LOCATION	COUNTY	LATITUDE	LONGITUDE
ECO66E09	Active	Clark Creek	06010108 Nolichucky	RM 1.8 National Forest property off Hwy 107 Clarks Creek Rd	Unicoi	36.15077	-82.52911
ECO66E11	Active	Lower Higgens Creek	06010108 Nolichucky	RM 1.7 Lower Higgins Cr Rd 1 mi NW Ernestville	Unicoi	36.08722	-82.52027
ECO66E17	Active	Double Branch	06010201 Fort Loudoun Lake	RM 0.1 U/S Millers Cove Rd	Blount	35.74378	-83.76631
ECO66E18	Active	Gee Creek	06020002 Hiwassee	RM 0.9 Near Gee Creek Wilderness Boundary	Polk	35.24444	-84.54388
ECO66F06	Active	Abrams Creek	06010204 Little Tennessee	RM 18.3 West end of Cades Cove, 0.6 mi U/S Mill Creek	Blount	35.59305	-83.84694
ECO66F07	Active	Beaverdam Creek	06010102 S Fork Holston	RM 5, 1 mi SW Backbone Rock Park	Johnson	36.58638	-81.8275
ECO66F08	Active	Stony Creek	06010103 Watauga	RM 12.5 U/S SR 91	Carter	36.46722	-81.99805
ECO66G04	Active	Middle Prong Little Pigeon R	06010107 Lower French Broad	RM 0.5 U/S restricted rd 0.2 mi east Greenbriar Cove	Sevier	35.70666	-83.37888
ECO66G05	Active	Little River	06010201 Ft Loudoun/Little R	RM 50.7 U/S last house Little River Trail above Elkmont	Sevier	35.65333	-83.57727
ECO66G07	Active	Citico Creek	06010204 Little Tennessee	RM 5.2, one mile U/S Jakes Creek	Monroe	35.50555	-84.10694
ECO66G09	Active	North River	06010204 Little Tennessee	RM 3, 500 meters U/S campground on North River Rd	Monroe	35.32777	-84.14583
ECO66G12	Active	Sheeds Creek	03150101 Conasauga	RM 1.8, 0.25 mi U/S Sheds Creek Rd	Polk	35.00305	-84.61222
ECO6701	Active	Big Creek	06010104 Holston	RM 9.8, D/S Fisher Creek West of Surgoinsville on Stanley Valley Rd	Hawkins	36.4778	-82.9387
ECO6702	Active	Fisher Creek	06010104 Holston	RM 0.6, U/S Bray Road	Hawkins	36.49	-82.94027
ECO6707	Probation	Possum Creek	06010102 South Fork Holston	RM 1.5, Weaver Pike Bridge, Bluff City	Sullivan	36.48	-82.19944
ECO67F06	Active	Clear Creek	06010207 Lower Clinch	RM 1, U/S Norris Municipal Park Road	Anderson	36.21361	-84.05972

ECOREGION REFERENCE STREAMS

SITE #	STATUS	STREAM	USGS HUC	LOCATION	COUNTY	LATITUDE	LONGITUDE
ECO67F13	Active	White Creek	06010205 Upper Clinch	RM 2, D/S old USGS gauging station next to White Creek Rd	Union	36.34361	-83.89166
ECO67F14	Active	Powell River	06010206 Powell	RM 106.5 McDowell Shoal D/S Fourmile Creek	Hancock	36.55638	-83.37916
ECO67F16	Active	Hardy Creek	06010206 Powell	RM 1.2, U/S SR 661 Powell Valley Rd	Lee County, VA	36.65083	-83.24722
ECO67F17	Active	Big War Creek	06010205 Upper Clinch	RM 0.6 Pawpaw Rd	Hancock	36.42694	-83.34694
ECO67F23	Active	Martin Creek	06010206 Powell	RM 0.5 Powell Valley Rd just U/S Hopkins Rd	Hancock	36.59111	-83.335
ECO67F25	Active	Powell River	06010206 Powell	RM 65.5 River Rd	Claiborne	36.55638	-83.60194
ECO67F27	Probation	Indian Creek	06010205 Upper Clinch	RM 3.7, off Indian Creek Road approx. 1 mi U/S Joe Mill Creek	Grainger	36.39519	-83.40339
ECO67G05	Active	Bent Creek	06010108 Nolichucky	RM 3.6 U/S Junction of Warrensburg and Mountain Rd	Hamblen	36.18793	-83.16414
ECO67G08	Probation	Brymer Creek	06020002 Hiwassee	RM 1.3, U/S Spring Br off Roark Lane/Brymer Creek Rd	Bradley	35.12666	-84.96388
ECO67G09	Active	Harris Creek	06020002 Hiwassee	RM 4.8, U/S Bancroft Rd	Bradley	35.175	-84.97916
ECO67G10	Active	Flat Creek	06010107 Lower French Broad	RM 12 D/S Muddy Hollow Rd	Sevier	35.9157	-83.4515
ECO67H06	Active	Laurel Creek	06010204 Little Tennessee	RM 0.8, D/S Laurel Creek Rd	Monroe	35.44829	-84.28833
ECO68A01	Active	Rock Creek	05130104 S Fork Cumberland	RM 24.8, Pickett State Park	Pickett	36.57833	-84.79472
ECO68A03	Active	Laurel Fork of Station Camp Cr	05130104 S Fork Cumberland	RM 4, Big South Fork NRRA	Fentress/ Scott	36.51611	-84.69805
ECO68A08	Active	Clear Creek	06010208 Emory	RM 4, Genesis Rd (HWY 298)	Morgan	36.11916	-84.7425
ECO68A13	Active	Piney Creek	06010201 Watts Bar Lake	RM 8.1, U/S Wash Pelfrey Rd, U/S Polecat Branch	Rhea	35.62083	-84.96944

ECOREGION REFERENCE STREAMS

SITE #	STATUS	STREAM	USGS HUC	LOCATION	COUNTY	LATITUDE	LONGITUDE
ECO68A20	Active	Mullens Creek	06020001 Tennessee	RM 5, U/S Jeep Trail	Marion	35.12472	-85.44388
ECO68A26	Active	Daddy's Creek	06010208 Emory	RM 2.3, U/S Hebbertsburg Rd, Catoosa	Cumberland	36.05861	-84.79138
ECO68A27	Probation	Island Creek	06010208 Emory	RM 2.3, U/S Noah Hambrey Rd, Catoosa	Morgan	36.05138	-84.66805
ECO68A28	Active	Rock Creek	06010208 Emory	RM 1.8, Off Hwy 62 approx 1 mi NE Lancing	Morgan	36.13277	-84.64166
ECO68B01	Active	Crystal Creek	06020004 Sequatchie	RM 1.2, Approx 0.25 mi D/S Lower East Valley Rd	Bledsoe	35.54083	-85.21694
ECO68B02	Active	McWilliams Creek	06020004 Sequatchie	RM 1.9, D/S Smith Rd	Sequatchie	35.4175	-85.32083
ECO68B09	Active	Mill Branch	06020004 Sequatchie	RM 0.4, U/S Upper East Valley Rd	Bledsoe	35.67444	-85.08888
ECO68C13	Active	Mud Creek	06030003 Upper Elk	RM 5.6, U/S E Roarks Cove Rd	Franklin	35.23055	-85.91722
ECO68C15	Active	Crow Creek	06030001 Guntersville Lake	RM 34.7, Off Ford Spring Rd below UT in Tom Pack Hollow	Franklin	35.1138	-85.9128
ECO68C20	Active	Crow Creek	06030001 Guntersville Lake	RM 35, Off Ford Spring Rd upstream UT in Tom Pack Hollow	Franklin	35.1155	-85.9110
ECO68C21	Active	Gilbreath Creek	06020001 Lower Tennessee	RM 0.1, Cove Loop Lower Road			
ECO69D03	Active	Flat Fork	06010208 Emory	RM 5, U/S Flat Fork Rd, U/S Rocky Fork Branch	Morgan	36.1235	-84.5122
ECO69D05	Active	New River	05140104 S Fork Cumberland	RM 55.4, approx 0.5 mi U/S HWY 116, 0.3 mi U/S Morgan/Anderson Co. line	Morgan	36.12444	-84.43130
ECO69D06	Probation	Round Rock Creek	05130104 S Fork Cumberland	RM 1, U/S ford off Norma Rd	Campbell	36.24722	-84.28444
ECO69E01	Active	No Business Branch	05130101 Upper Cumberland	RM 0.2, U/S Hwy 25	Campbell	36.55277	-84.06861
ECO69E04	Active	Stinking Creek	05130101 Upper Cumberland	RM 15.1, Approx 0.5 mi south of Stinking Creek Rd near power line	Campbell	36.4258	-84.2618

ECOREGION REFERENCE STREAMS

SITE #	STATUS	STREAM	USGS HUC	LOCATION	COUNTY	LATITUDE	LONGITUDE
ECO71E09	Active	Buzzard Creek	05130206 Red	RM 1.3, Buzzard Creek Rd	Robertson	36.60583	-86.98361
ECO71E14	Active	Passenger Creek	05130206 Red	RM 1.6, HWY 76	Montgomery	36.53444	-87.19583
ECO71E17	Probation	Brush Creek	05130206 Red	RM 5, Stroudville Rd	Robertson	36.481389	-87.089722
ECO71E18	Active	Santee Creek	05130206 Red	RM 0.9, Sprouse Rd	Robertson	36.49778	-86.778333
ECO71E19	Active	Calebs Creek	05130206 Red	RM 1.2, U/S Maxie/Carr Rd	Robertson	36.49237	-87.0066
ECO71F12	Active	South Harpeth Creek	05130204 Harpeth	RM 16.9, South Harpeth and Pewitt Rd, U/S Kelley Creek	Williamson	35.925	-87.0929
ECO71F16	Active	Wolf Creek	06040003 Lower Duck	RM 0.8, U/S Wolf Creek Rd	Hickman	35.81805	-87.68527
ECO71F19	Active	Brush Creek	06040004 Buffalo	RM 2.1, Paul Reed Rd, D/S Little Brush Creek	Lewis/Lawrence	35.41972	-87.53416
ECO71F27	Active	Swanegan Branch	06030005 Pickwick Lake	RM 0.5, Off Thomas Woodard Rd	Wayne	35.06916	-87.6375
ECO71F28	Active	Little Swan Creek	06040003 Lower Duck	RM 5.6, Meriwether Lewis National Monument	Lewis	35.52888	-87.45361
ECO71F29	Active (2/27/03)	Hurricane Creek	06040003 Lower Duck	RM 7.6, Off Vaden Branch Road	Humphreys	35.980556	-87.761389
ECO71G03	Active	Flat Creek	05130106 Upper Cumberland	RM 1.8, Hwy 136	Putnam	36.35944	-85.43138
ECO71G04	Active	Spring Creek	05130106 Upper Cumberland	RM 16.2, Boatman Rd	Overton	36.27277	-85.42333
ECO71G10	Active	Hurricane Creek	06030003 Upper Elk	RM 9.4, Hurricane Creek Rd	Moore	35.32083	-86.29944
ECO71G14	Probation	Blackburn Fork	05130106 Upper Cumberland	RM 14.5, Cummins Mill Rd	Jackson	36.2506	-85.5647
ECO71G16	PROBATION	Line Creek	05110002 Barren	RM 13.9, Clementsville KY Rd (Clementsville Church of Christ)	CLAY	36.6106	-85.7258

ECOREGION REFERENCE STREAMS

SITE #	STATUS	STREAM	USGS HUC	LOCATION	COUNTY	LATITUDE	LONGITUDE
ECO71H03	Active	Flynn Creek	05130106 Upper Cumberland	RM 10.2, Flynn Creek Rd, 3 mi NE Nameless TN	Jackson	36.2792	-85.66444
ECO71H06	Active	Clear Fork	05130108 Caney Fork	RM 6.8, Off Big Hill Rd	DeKalb/Cannon	35.92416	-85.99083
ECO71H09	Active	Carson Fork	05130203 Stones	RM 4.2, Burt-Burgen Rd, 2 mi NE Bradyville	Cannon	35.76495	-86.13263
ECO71I10	Active	Flat Creek	06040002 Upper Duck	RM 6.4, U/S Hazelwood Rd	Marshall	35.68583	-86.80166
ECO71I12	Active	Cedar Creek	05130201 Cumberland	RM 4.6, Centerville Rd	Wilson	36.28425	-86.20339
ECO71I14	Active	Little Flat Creek	06040002 Upper Duck	RM 3.6, U/S Will Brown Rd	Maury	35.69903	-86.83872
ECO71I15	Active	Harpeth River	05130204 Harpeth	RM 106.4, D/S McDaniel Rd	Williamson	35.8325	-86.70019
ECO71I16	Active	West Fork Stones River	05130203 Stones	RM 30.4, Walnut Grove Rd	Rutherford	35.7225	-86.4451
ECO71I17	New	Spring Creek	5130201 Old Hickory Res	RM19.2, Eastover Rd	Wilson	36.179999	-86.241111
ECO73A01	Active	Cold Creek	08010100 Mississippi	RM 14.4, U/S Long Hole Rd	Lauderdale	35.7425	-89.6994
ECO73A02	Active	Middle Fork Forked Deer	08010100 Mississippi	RM 3.3, 0.5 miles upstream Watkins Rd	Lauderdale	35.81777	-89.65611
ECO73A03	Active	Cold Creek	08010100 Mississippi	RM 2.3, Approx 1.4 mi u/s Crutcher Lake Rd, U/S Adams Bayou	Lauderdale	35.66305	-89.81222
ECO73A04	Active	Bayou du Chien	08010202 Obion	RM 3.2, Approx 1.5 mi U/S boat ramp on Walnut Log Rd and 0.75 mi U/S last cabin	Lake	36.475	-89.30916
ECO74A06	Active	Sugar Creek	08010100 Mississippi	RM 2.3, U/S Copper Rd	Tipton	35.49944	-89.91914
ECO74A08	Active	Pawpaw Creek	08010202 Obion	RM 3.1, U/S Upper Crossing of Putnam Hill Rd	Obion	36.30527	-89.35666
ECO74B01	Active	Terrapin Creek	08010202 Obion	RM 1.6, Terrapin Creek Rd	Henry	36.48666	-88.48583

ECOREGION REFERENCE STREAMS

SITE #	STATUS	STREAM	USGS HUC	LOCATION	COUNTY	LATITUDE	LONGITUDE
ECO74B04	Active	Powell Creek	08010202 Obion	RM 2.2, McClains Levee Rd	Weakley	36.48027	-88.64
ECO74B12	Active	Wolf River	08010210 Wolf	RM 72.7, U/S Yager Rd	Fayette	35.0325	-89.24583
ECO74B12A	Active	Wolf River	08010210 Wolf	D/S Yager Road	Fayette	35.03262	-89.24862
FECO65E03	Active	Unnamed Tributary to Unnamed tributary to Dabbs Creek	06040001 Tennessee Western Valley	RM 0.1, Natchez Trace State Forest Off Todd Trail	Henderson	35.79006	-88.30636
FECO65E04	Active	Unnamed tributary to Cub Creek	06040001 Tennessee Western Valley	RM 0.1, Natchez Trace State Park Off Taylor Trail	Henderson	35.78489	-88.26502
FECO65E05	New	Tuscumbia River UT	08010207	RM 0.6, Big Hill State Park @ Footbridge on Tuscambia Bend Trail		35.05162	-88.74677
FECO65J01	New	Haw Br	06030005 TN-Pickwick Lake	RM 0.9, U/S Pickwick Embayment	Hardin	35.0852	-88.1916
FECO65J02	Active	Unnamed tributary to Horse Creek	06040001 Tennessee Western Valley	RM 0.3, Sugar Camp Hollow	Hardin	35.15521	-88.19176
FECO65J03	Active	English Creek	06040001 Tennessee Western Valley	RM 5.6, South Of Firetower Road Near Seaton Cabin And Ross Property Line	Hardin	35.15393	-88.17444
FECO66D01	Active	Black Branch	06010103 Watauga	RM 2 Hwy 321 near Elk Mills	Carter	36.2825	-82.0275
FECO66D06	Active	Tumbling Creek	06010108 Nolichucky	RM 1.5 Tumbling Creek Rd end	Carter	36.01805	-82.48194
FECO66D07	Active	Little Stoney Creek	06010103 Watauga	RM 2 Little Stony Rd 0.3 mi D/S Goodwin Field Br	Carter	36.28666	-82.06666
FECO66G01	Active	Indian Branch	06010204 Little Tennessee	RM 0.1North River Rd	Monroe	35.33102	-84.06733
FECO67F02	Active	Mill Creek	06010207 Lower Clinch	Off Cave Road	Roane	35.84999	-84.38210

ECOREGION REFERENCE STREAMS

SITE #	STATUS	STREAM	USGS HUC	LOCATION	COUNTY	LATITUDE	LONGITUDE
FECO67G11	Active	North Prong Fishdam Creek	06010104 Holston	RM 1.6, U/S SR 34	Sullivan	36.5344	-82.0192
FECO67H04	Active	Blackburn Creek	06020002 Hiwassee	RM1.8, 0.24 mi U/S Blackburn Hollow Rd	Bradley	35.22472	-84.97055
FECO67H08	Active	Parker Creek	06010104 Holston	RM 0.5, Holston Army Ammunition Property	Hawkins	36.5225	-82.65888
FECO67I12	Active	Mill Branch	06010207 Lower Clinch	RM 1.2, Below confluence of 2 tribs off Tuskegee Drive	Anderson	35.98833	-84.28888
FECO68A01	Active	Douglas Branch	06010208 Emory	RM 0.1, Barnett Bridge Road	Morgan	36.1	-84.777
FECO68C12	Active	Ellis Gap Branch	06020001 Tennessee	RM 0.4, U/S Mullens Cove Rd, Prentice Cooper State Forest	Marion	35.04916	-85.47277
FECO69D01	Active	Unnamed tributary to New River	05130104 South Fork Cumberland	RM 0.1, U/S Hwy 116	Morgan	36.12090	-84.43214
FECO69D03	New	Bear Branch	06010205 Upper Clinch	RM 0.1, U/S Hwy 68	Campbell	36.39916	-84.30928
FECO69D04	New	Wheeler Creek UT	05130104 Cumberland South Fork	RM 0.6, @ Big Bruce Ridge	Campbell	36.30771	-84.27522
FECO71E01	New	Sulphur Fork UT	05130206 Red	RM 0.1, OFF HWY 256	Robertson	36.51460	-87.05695
FECO71E02	New	Savage Branch	05130206 Red	RM 1.2, U/S Distillery Rd Off Hwy 76	Robertson	36.47534	-86.76083
FECO71E03	New	Brush Creek	05130206 Red	Rm 9.0, U/S Gause Rd	Robertson	36.4342	-87.06622
FECO71F01	Active	Unnamed tributary to Little Swan Creek	06040003 Lower Duck	RM 0.1, Off DP Humphreys Rd	Lewis	35.5	-87.418
FECO71F02	New	Hurricane Creek UT	06040004 Buffalo	U/S Hurricane Creek Rd (Bryson Hollow)		35.57647	-87.75973
FECO71F03	Active	Ethridge Hollow	06040003 Lower Duck	RM 0.1, U/S Hwy 230	Humphreys	35.9407	-87.6530
FECO71F04	New	Marrowbone Creek UT	05130202 Cumberland (Cheatham)	RM 0.1, U/S Little Morrowbone Rd in Beamsn City park	Davidson	36.27212	-86.9049
FECO71G01	Active	Flat Creek	05130106 Upper Cumberland	RM 8.3, Upper Hillman Rd	Overton	36.4	-85.374

ECOREGION REFERENCE STREAMS

SITE #	STATUS	STREAM	USGS HUC	LOCATION	COUNTY	LATITUDE	LONGITUDE
FECO71G02	New	Long Fork UT	05110002 Barren	RM 0.1, U/S Tanyard Rd	Macon	36.48909	-85.93973
FECO71H03	New	Haws Spring FK	05130203 Stones	RM 2.7, off farm rd off Jimtown Rd	Cannon	35.761291	-86.08854
FECO71I02	New	Young Branch	5130201 Cumberland (Old Hickory)	RM 1.6, U/S Hwy 70N	Wilson	36.24031	-86.16099
FECO71I03	New	McKnight Branch UT	05130203 Stones	RM 2.4, U/S Ford off Elrod McElroy	Rutherford	35.896901	-86.18094
FECO71I04	New	East Fork Hurricane Ck	05130203 Stones	RM 2.2, Cedar Forest Rd in Cedars of Lebanon State Park	Wilson	36.05598	-86.27829
FECO71I05	New	West Fk Stones River	05130203 Stones	RM 37.6, U/S Harrison Rd	Rutherford	35.65667	-86.45599
FECO74A01	Active	Unnamed tributary to Pawpaw Creek	08010202 Obion	RM 0.4, Off Putnam Hill Rd	Obion	36.31379	-89.34322
FECO74B01	Active	Unnamed tributary to North Fork Wolf River	08010210 Wolf	RM 0.2, Ames Plantation	Fayette	35.10770	-89.31641
FECO74B02	New	Hatchie River UT	08010208 Lower Hatchie	RM 2.7, off Landfill Rd	Haywood	35.54557	-89.30765

Regional Expectations for Individual Habitat Parameters

	Epifaunal Substrate		Embedded-ness		Pool Substrate		Velocity Depth		Pool Variability		Sediment Deposition		Flow Status		Channel Alteration		Riffle Frequency		Channel Sinuosity		Bank Stability		Vegetative Protection		Riparian Vegetation	
Eco	REF	75%	REF	75%	REF	75%	REF	75%	REF	75%	REF	75%	REF	75%	REF	75%	REF	75%	REF	75%	REF	75%	REF	75%	REF	75 %
65a	5	4	NA	NA	7	5	NA	NA	11	8	12	9	11	8	12	9	NA	NA	5	4	8	6	14	10	4	3
65b	15	11	NA	NA	10	7	NA	NA	10	8	9	7	10	8	16	12	NA	NA	12	9	13	10	19	14	20	15
65e	15	11	NA	NA	11	8	NA	NA	11	8	15	11	18	14	18	14	NA	NA	15	11	16	12	20	15	18	14
65i	15	11	14	10	10	8	NA	NA	12	9	12	9	8	6	18	14	8	6	11	8	10	8	18	14	20	15
65j	17	13	18	14	NA	NA	17	13	NA	NA	16	12	15	11	19	14	18	14	NA	NA	18	14	20	15	20	15
66d	20	15	20	15	NA	NA	20	15	NA	NA	18	14	19	14	20	15	20	15	NA	NA	20	15	20	15	20	15
66e	19	14	18	14	NA	NA	20	15	NA	NA	18	14	18	14	20	15	20	15	NA	NA	20	15	20	15	20	15
66f	18	14	19	14	NA	NA	16	12	NA	NA	19	14	19	14	20	15	18	14	NA	NA	20	15	20	15	18	14
66g	19	14	19	14	NA	NA	18	14	NA	NA	19	14	18	14	20	15	20	15	NA	NA	20	15	20	15	19	14
67f	18	13	18	14	NA	NA	17	13	NA	NA	15	11	18	14	20	15	19	14	NA	NA	18	14	19	14	20	15
67g	16	12	15	11	NA	NA	16	11	NA	NA	15	11	17	13	15	11	16	12	NA	NA	12	9	16	12	12	9
67h	13	10	17	13	NA	NA	16	12	NA	NA	15	11	15	11	18	13	18	14	NA	NA	18	14	19	14	19	14
67i	13	10	16	12	NA	NA	11	8	NA	NA	15	11	17	13	16	12	13	10	NA	NA	18	14	19	14	16	12
68a Jan-Jun	18	14	18	14	NA	NA	17	13	NA	NA	19	14	19	14	19	14	18	14	NA	NA	20	15	20	15	20	15
68a Jul-Dec	18	13	17	13	NA	NA	15	11	NA	NA	18	14	15	11	19	14	14	11	NA	NA	20	15	20	15	20	15
68b Jan-Jun	15	11	16	12	NA	NA	15	12	NA	NA	13	10	17	13	18	13	16	12	NA	NA	14	10	16	12	13	10
68b Jul-Dec	14	10	12	9	NA	NA	12	9	NA	NA	10	7	15	11	16	12	16	12	NA	NA	14	11	13	10	11	8
68c Jan-Jun	17	13	16	12	NA	NA	15	11	NA	NA	17	13	18	14	19	14	18	14	NA	NA	17	13	19	14	18	14
68c Jul-Dec	16	12	16	12	NA	NA	14	10	NA	NA	16	12	12	9	20	15	18	13	NA	NA	16	12	18	14	18	13

	Epifaunal Substrate		Embedded-ness		Pool Substrate		Velocity Depth		Pool Variability		Sediment Deposition		Flow Status		Channel Alteration		Riffle Frequency		Channel Sinuosity		Bank Stability		Vegetative Protection		Riparian Vegetation	
Eco	REF	75%	REF	75%	REF	75%	REF	75%	REF	75%	REF	75%	REF	Eco	REF	75%	REF	75%	REF	75%	REF	75%	REF	75%	REF	75 %
69d Jan-Jun	17	13	18	14	NA	NA	18	14	NA	NA	18	13	16	12	20	15	19	14	NA	NA	19	14	20	15	20	15
69d Jul-Dec	16	12	17	13	NA	NA	15	11	NA	NA	17	13	9	7	20	15	18	14	NA	NA	18	14	20	15	20	15
71e	16	12	15	11	NA	NA	16	12	NA	NA	13	10	16	12	17	13	16	12	NA	NA	13	10	14	11	13	10
71f	16	12	17	13	NA	NA	16	12	NA	NA	15	11	15	11	18	14	17	13	NA	NA	16	12	18	14	16	12
71g	15	11	16	12	NA	NA	16	12	NA	NA	16	12	17	13	18	14	17	13	NA	NA	18	14	18	14	16	12
71h	15	11	17	13	NA	NA	16	12	NA	NA	15	11	16	12	16	12	17	13	NA	NA	17	13	15	11	12	9
71i Jan-Jun	12	9	14	10	15	11	14	10	13	10	14	10	16	12	17	12	11	8	16	12	16	12	16	12	14	10
71i Jul-Dec	13	10	13	10	11	8	10	8	8	6	13	10	12	9	18	14	8	6	9	7	16	12	16	12	13	9
71I Jul-Dec	13	10	13	10	11	8	10	8	8	6	13	10	12	9	18	14	8	6	9	7	16	12	16	12	13	9
73a	11	8	NA	NA	8	6	NA	NA	6	5	9	7	15	11	17	13	NA	NA	10	8	10	8	17	13	18	14
74a	10	8	13	10	NA	NA	13	10	NA	NA	9	8	8	6	15	11	14	10	NA	NA	11	8	12	9	13	10
74b	12	9	NA	NA	11	8	NA	NA	10	8	10	7	15	11	16	12	NA	NA	10	8	13	10	20	15	20	15

APPENDIX B

FORMS AND DATA SHEETS

COUNTY AND STATE ABBREVIATIONS AND CODE NUMBERS
HABITAT ASSESSMENT DATA SHEETS
RAPID PERIPHYTON SURVEY (RPS) DATA SHEET
BIOLOGICAL SAMPLE REQUEST INCLUDING CHAIN OF CUSTODY FORM
NON-DIATOM ALGAE TAXONOMIC BENCH SHEET
DIATOM ALGAE TAXONOMIC BENCH SHEET
PERIPHYTON ASSESSMENT REPORT

COUNTY AND STATE – Abbreviations and Code Numbers

COUNTY NAME	WPC CO ABBR	TN CO NO	NATIONAL TN FIPS	COUNTY NAME	WPC CO ABBR	TN CO NO	NATIONAL TN FIPS
ANDERSON	**AN**	01	001	LAUDERDALE	**LE**	49	097
BEDFORD	**BE**	02	003	LAWRENCE	**LW**	50	099
BENTON	**BN**	03	005	LEWIS	**LS**	51	101
BLEDSOE	**BL**	04	007	LINCOLN	**LI**	52	103
BLOUNT	**BT**	05	009	LOUDON	**LO**	53	105
BRADLEY	**BR**	06	011	MCMINN	**MM**	54	107
CAMPBELL	**CA**	07	013	MCNAIRY	**MC**	55	109
CANNON	**CN**	08	015	MACON	**MA**	56	111
CARROLL	**CR**	09	017	MADISON	**MN**	57	113
CARTER	**CT**	10	019	MARION	**MI**	58	115
CHEATHAM	**CH**	11	021	MARSHALL	**ML**	59	117
CHESTER	**CS**	12	023	MAURY	**MY**	60	119
CLAIBORNE	**CL**	13	025	MEIGS	**ME**	61	121
CLAY	**CY**	14	027	MONROE	**MO**	62	123
COCKE	**CO**	15	029	MONTGOMERY	**MT**	63	125
COFFEE	**CE**	16	031	MOORE	**MR**	64	127
CROCKETT	**CK**	17	033	MORGAN	**MG**	65	129
CUMBERLAND	**CU**	18	035	OBION	**OB**	66	131
DAVIDSON	**DA**	19	037	OVERTON	**OV**	67	133
DECATUR	**DE**	20	039	PERRY	**PE**	68	135
DE KALB	**DB**	21	041	PICKETT	**PI**	69	137
DICKSON	**DI**	22	043	POLK	**PO**	70	139
DYER	**DY**	23	045	PUTNAM	**PU**	71	141
FAYETTE	**FA**	24	047	RHEA	**RH**	72	143
FENTRESS	**FE**	25	049	ROANE	**RO**	73	145
FRANKLIN	**FR**	26	051	ROBERTSON	**RN**	74	147
GIBSON	**GI**	27	053	RUTHERFORD	**RU**	75	149
GILES	**GS**	28	055	SCOTT	**SC**	76	151
GRAINGER	**GR**	29	057	SEQUATCHIE	**SE**	77	153
GREENE	**GE**	30	059	SEVIER	**SV**	78	155
GRUNDY	**GY**	31	061	SHELBY	**SH**	79	157
HAMBLEN	**HA**	32	063	SMITH	**SM**	80	159
HAMILTON	**HM**	33	065	STEWART	**ST**	81	161
HANCOCK	**HK**	34	067	SULLIVAN	**SU**	82	163
HARDEMAN	**HR**	35	069	SUMNER	**SR**	83	165
HARDIN	**HD**	36	071	TIPTON	**TI**	84	167
HAWKINS	**HS**	37	073	TROUSDALE	**TR**	85	169
HAYWOOD	**HY**	38	075	UNICOI	**UC**	86	171

COUNTY NAME	WPC CO ABBR	TN CO NO	NATIONAL TN FIPS	COUNTY NAME	WPC CO ABBR	TN CO NO	NATIONAL TN FIPS
HENDERSON	**HE**	39	077	UNION	**UN**	87	173
HENRY	**HN**	40	079	VAN BUREN	**VA**	88	175
HICKMAN	**HI**	41	081	WARREN	**WA**	89	177
HOUSTON	**HO**	42	083	WASHINGTON	**WN**	90	179
HUMPHREYS	**HU**	43	085	WAYNE	**WE**	91	181
JACKSON	**JA**	44	087	WEAKLEY	**WY**	92	183
JEFFERSON	**JE**	45	089	WHITE	**WH**	93	185
JOHNSON	**JO**	46	091	WILLIAMSON	**WI**	94	187
KNOX	**KN**	47	093	WILSON	**WS**	95	189
LAKE	**LA**	48	095				
STATE NAME	**WPC ABBR**			**STATE NAME**	**WPC ABBR**		
ALABAMA	**_AL**			MISSISSIPPI	**_MS**		
ARKANSAS	**_AR**			MISSOURI	**_MO**		
GEORGIA	**_GA**			N. CAROLINA	**_NC**		
KENTUCKY	**_KY**			VIRGINIA	**_VA**		

HABITAT ASSESSMENT DATA SHEET- HIGH GRADIENT STREAMS (FRONT)

STREAM NAME	LOCATION
STATION #_____	ECOREGION
LAT_____LONG_____	WATERSHED GROUP
WBID/HUC	INVESTIGATORS
FORM COMPLETED BY	DATE_____ TIME_____

	Optimal	Suboptimal	Marginal	Poor
1. Epifaunal Substrate/Available Cover	Over 70% of stream reach has natural stable habitat suitable for colonization by fish and/or macroinvertebrates. Four or more productive habitats are present.	Natural stable habitrat covers 40-70% of stream reach. Three or more productive habitats present. (If near 70% and more than 3 go to optimal)	Natural stable habitat covers 20 -40% of stream reach. Two or more productive habitats present. (If near 40% and more than 2 go to suboptimal)	Less than 20% stable habitat; lack of habitat is obvious; substrate unstable or lacking.
SCORE	20 19 18 17 16	15 14 13 12 11	10 9 8 7 6	5 4 3 2 1
Comments				
2. Embeddedness of Riffles	Gravel, cobble, and boulder s are 0-25% surrounded by fine sediment. Layering of cobble provides diversity of niche space. If near 25% drop to suboptimal if riffle is not layerd cobble..	Gravel, cobble and boulders are 25-50% surrounded by fine sediment. Niches in slower areas of riffle and in bottom layers of cobble have become compromised. If nearing 50% and riffles are not layered cobble drop to marginal.	Gravel, cobble, and boulder s are 50-75% surrounded by fine sediment. Nich space in middle layers of cobble is starting to fill with fine sediment.	Gravel, cobble, and boulder s are more than 75% surrounded by fine sediment. Niche space is reduced to a single layer or is absent.
SCORE	20 19 18 17 16	15 14 13 12 11	10 9 8 7 6	5 4 3 2 1
Comments				
3. Velocity/Depth Regime	All four velocity/depth regimes present (slow-deep, slow-shallow, fast-deep, fast-shallow)	Only 3 of the 4 regimes present (if fast-shallow is missing score lower). If slow-deep missing score 15.	Only 2 of the 4 habitat regimes present (if fast-shallow or slow-shallow are missing, score low)	Dominated by 1 velocity/depth regime. Others reimes too small or infrequent to support aquatic populations.
SCORE	20 19 18 17 16	15 14 13 12 11	10 9 8 7 6	5 4 3 2 1
Comments				
4. Sediment Deposition	Sediment deposition affects less than 5% of stream bottom in quiet areas. New deposition on islands and point bars is absent or minimal.	Sediment deposition affects 5-30% of stream bottom. Slight deposition in pool or slow areas. Some new deposition on islands and point bars. Move to marginal if build-up approaches 30%..	Sediment deposition affects 30-50% of stream bottom. Sediment deposits at obstruction, constrictions and bends. Moderate pool deposition.	Heavy deposits of fine material, increased bar development; more than 50% of the bottom changing frequently; pools almost absent due to substantial sediment deposition
SCORE	20 19 18 17 16	15 14 13 12 11	10 9 8 7 6	5 4 3 2 1
Comments				
5. Channel Flow Status. Do not evaluate in area of reach that is backed up by obstructions.	Water reaches base of both lower banks, and minimal amount of productive habitat is exposed.	Water fills> 75% of the available channel; or 25 % of productive habitat is exposed.	Waters fills 25-75 % of the available channel, and/or stable habitat is mostly exposed.	Very little water in channel and mostly present as standing pools.. Little or no productive habitat due to lack of water.
Comments				
SCORE	20 19 18 17 16	15 14 13 12 11	10 9 8 7 6	5 4 3 2 1

HABITAT ASSESSMENT DATA SHEET- HIGH GRADIENT STREAMS (BACK)

	Optimal	**Suboptimal**	**Marginal**	**Poor**
6. Channel Alteration	Channelization , dredging or 4-wheel activity absent or minimal; stream with natural meander pattern. NO bridges, culverts, shoring or artificial structures in reach. Upstream or downstream structures do no affect reach	Channelization, dredging or 4-wheel activity up to 40%. Channel has stabilized. NO bridges, culverts, shoring or artificial structures in reach. Upstream or downstream structures do not affect reach.	Channelization, dredging or 4-wheel activity 40-80% or any amount of channelization that has not stabilized. Bridges, culverts, shoring or other artificial structures may be within reach. Upstream or downstream structures may have affected flow pattern.	Over 80% of the stream reach channelized, dredged or affected by 4-wheelers. Instream habitat greatly altered or removed. Shoring structures may be common. Artificial structures upstream or downstream of reach may have greatly affected flow patterns in reach.
Comments				
SCORE	20 19 18 17 16	15 14 13 12 11	10 9 8 7 6	5 4 3 2 1
7. Frequency of riffles, bends or other re-oxygenation zones. Use frequency for category. Rank by quality.	Occurrence of re-oxygenation zones relatively frequent; ratio of distance between areas divided by width of the stream <7:1.	Occurrence of re-oxygenation zones infrequent; distance between areas divided by the width of the stream is between 7 to 15.	Occasional re-oxygenation area. The distance between areasdivided by the width of the stream is over 15 and up to 25.	Generally all flat water or flat bedrock little opportunity for re-oxugenation. Distance between areas divided by the width of the stream is a ratio of >25.
Comments				
SCORE	20 19 18 17 16	15 14 13 12 11	10 9 8 7 6	5 4 3 2 1
8. Bank Stability (score each bank) Determine left or right side by facing downstream.	Banks stable; evidence of erosion or bank failure absent or minimal; little potential for future problems <5% of bank affected.	Moderately stable; infrequent, small areas of erosion mostly healed over. 5-30% of bank in reach has areas of erosion. If approaching 30% score marginal if banks steep.	Moderately unstable; 30-60 % of bank in reach has areas of erosion; high erosion potential during floods, If approaching 60% score poor if banks steep.	Unstable; many eroded area; raw areas frequent along straight sections and bends; obvious bank sloughing; 60-100% of bank has erosional scars
Commments				
SCORE____(LB)	Left Bank 10 9	8 7 6	5 4 3	2 1 0
SCORE____(RB)	Right Bank 10 9	8 7 6	5 4 3	2 1 0
9. Vegetative Protective (score each bank) includes vegetation from top of bank to base of bank. Determine left or right side by facing downstream	More than 90% of the bank covered by undisturbed native vegetation, All 4 classes (mature trees, understory trees, shrubs, groundcover) are represented and allowed to grow naturally.	70-90% of the bank covered by native vegetation, If higher, than one class not well represented. Disruption evident but not effecting full plant growth.	50-70% of the bank covered by native vegetation. If more than two classes of vegetation missing. Non-native vegetation or closely cropped vegetation may be common.	Less than 50% of the bank covered by native vegetation or more than 2 classesare not well represented or most vegetation has been cropped..
Comments				
SCORE____(LB)	Left Bank 10 9	8 7 6	5 4 3	2 1 0
SCORE____(RB)	Right Bank 10 9	8 7 6	5 4 3	2 1 0
10. Riparian Vegetative Zone Width (score each bank. Zone begins at top of bank.	Width of riparian zone > 18 meters throughout reach. Unpaved footpaths may score 9 if run-off potential is neglible.	Width of riparian zone 12-18 meters throughout reach. Score high if areas < 18 meters are small or are minimally disturbed.	Width of riparian zone 6-11 meters throughout reach. Score high if areas less than 12 meters are small or are minimally disturbed.	Width of riparian zone <6 meters. Score high if areas less than 6 meters are small or are minimally disturbed.
Comments				
SCORE____(LB)	Left Bank 10 9	8 7 6	5 4 3	2 1 0
SCORE____(RB)	Right Bank 10 9	8 7 6	5 4 3	2 1 0

TOTAL SCORE _____ **Comparison to Ecoregional Guidelines** _____

If Score low, result of: _____**Natural Conditions or** _____ **Human Disturbance**

HABITAT ASSESSMENT DATA SHEET- LOW GRADIENT STREAMS (FRONT)

STREAM NAME	LOCATION
STATION #_____	ECOREGION
LAT_____ LONG_____	WATERSHED GROUP
WBID/HUC	INVESTIGATORS
FORM COMPLETED BY	DATE_____ TIME_____

	Optimal	Suboptimal	Marginal	Poor
1. Epifaunal Substrate/Available Cover	Greater than 50% of substrate favorable for macroinvertebrate and/or fish colonization Mix of three or more stable habitats present (undercut rooted banks, snags, macrophytes etc.)	30-50% mix of stable habitat; well-suited for full colonization potential; At leaset one typical habitat is missing.. If nearing 30% and more than 1 missing drop to marginal.	10-30% mix of stable habitat; availability less than desirable; substrate frequently disturbed or removed Habitat diversity is reduced, one or more habitat is missing while at least one other is inadequate.	Less than 10% stable habitat; lack of habitat is obvious; substrate unstable or lacking
Comments				
SCORE	20 19 18 17 16	15 14 13 12 11	10 9 8 7 6	5 4 3 2 1
2. Channel Substrate Characterization	Good mixture of substrate materials, with gravel and firm sand prevalent; root mats and submerged vegetation common	Mixture of soft sand, mud, or clay; mud may be dominant; some root mats and submerged vegetation present.	All mud or clay or soft sand bottom; little or no root mat; no submerged vegetation present.	Hard-pan clay, conglomerate or bedrock; no root mat or vegetation.
Comments				
SCORE	20 19 18 17 16	15 14 13 12 11	10 9 8 7 6	5 4 3 2 1
3. Pool Variability	Even mix of large-shallow, large-deep, small-shallow, small-deep pools present.	Majority of pools large-deep; very few shallow.	Shallow pools much more prevalent than deep pools.	Majority of pools small-shallow or pools absent.
Comments				
SCORE	20 19 18 17 16	15 14 13 12 11	10 9 8 7 6	5 4 3 2 1
4. Sediment Deposition	Little or no enlargement of islands or point bars and less than 20% of the bottom affected by sediment deposition	Some new increase in bar formation, mostly from gravel, sand or fine sediment 20-50% of bottom affected. Slight deposition in pools.	Moderate deposition of new gravel, sand or fine sediment on old and new bars 50-80% of bottom affected; sediment deposits at obstructions, constrictions, and bends; moderate deposition of pools	Heavy deposits of fine material, increased bar development; more than 80% of the bottom changing frequently; pools almost absent due to substantial sediment deposition
Comments				
SCORE	20 19 18 17 16	15 14 13 12 11	10 9 8 7 6	5 4 3 2 1
5. Channel Flow Status Do not evaluate in area of reach that is backed up by obstructions.	Water reaches base of both lower banks, and minimal amount productive habitat is exposed.	Water fills> 75% of the available channel; or 25 % of productive habitat is exposed.	Waters fills 25-75 % of the available channel, and/or productive habitat is mostly exposed.	Very little water in channel and mostly present as standing pools.. Little or no productive habitat available. due to lack of flow..
Comments				
SCORE	20 19 18 17 16	15 14 13 12 11	10 9 8 7 6	5 4 3 2 1

HABITAT ASSESSMENT DATA SHEET- LOW GRADIENT STREAMS (BACK)

	Optimal	**Suboptimal**	**Marginal**	**Poor**
6. Channel Alteration	Channelization, 4-wheel activity or dredging absent or minimal; stream with normal pattern. Shoring structures absent. Artifical structures absent in reach, structures upstream or downstream do not affect reach..	Channelization, 4-wheelingi or dtredging up to 40% of reach. Past channelization, has stabilized. Artifical structures absent, structures upstream or downstream do not affect reach.	Channelization, 4-wheelers or dredging 40 to 80% of stream or less that has not stabilized. Artificial structures may be in reach. Structures upstream or downstream may have affected flow pattern.	Over 80% of the stream reach channelized, dredged, shored or affected by 4-wheel activity. Instream habitat greatly altered or removed entirely. Artificial structures in or out of reach may have greatly affected flow..
Comments				
SCORE	20 19 18 17 16	15 14 13 12 11	10 9 8 7 6	5 4 3 2 1
7. Channel Sinuosity (Entire meander sequence not limited to sampling reach).	The bends in the stream increase the stream length 3-4 times longer than if it was in a straight line	The bends in the stream increase the stream length 2-3 times longer than if it was in a straight line.	The bends in the stream increase the stream length 2 to 1 times longer than if it was in a straight line.	Channel straight; waterway has been channelized for a long distance.
Comments				
SCORE	20 19 18 17 16	15 14 13 12 11	10 9 8 7 6	5 4 3 2 1
8. Bank Stability (score each bank) Determine left or right side by facing downstream.	Banks stable; evidence of erosion or bank failure absent or minimal; little potential for future problems <5% of bank affected.	Moderately stable; infrequent, small areas of erosion mostly healed over. 5-30% of bank in reach has areas of erosion. If approaching 30% score marginal if banks steep.	Moderately unstable; 30-60 % of bank in reach has areas of erosion; high erosion potential during floods, If approaching 60% score poor if banks steep.	Unstable; many eroded area; raw areas frequent along straight sections and bends; obvious bank sloughing; 60-100% of bank has erosional scars
Comments				
SCORE____(LB)	Left Bank 10 9	8 7 6	5 4 3	2 1 0
SCORE____(RB)	Right Bank 10 9	8 7 6	5 4 3	2 1 0
9. Vegetative Protective (score each bank) includes vegetation from top of bank to base of bank. Determine left or right side by facing downstream	More than 90% of the bank covered by undisturbed native vegetation, All 4 classes (mature trees, understory trees, shrubs, groundcover) are represented and allowed to grow naturally.	70-90% of the bank covered by native vegetation, If higher, than one class not well represented. Disruption evident but not effecting full plant growth.	50-70% of the bank covered by native vegetation. If more than two classes of vegetation missing. Non-native vegetation or closely cropped vegetation may be common.	Less than 50% of the bank covered by native vegetation or more than 2 classesare not well represented or most vegetation has been cropped..
Comments				
SCORE____(LB)	Left Bank 10 9	8 7 6	5 4 3	2 1 0
SCORE____(RB)	Right Bank 10 9	8 7 6	5 4 3	2 1 0
10. Riparian Vegetative Zone Width (score each bank. Zone begins at top of bank.	Width of riparian zone > 18 meters throughout reach. Unpaved footpaths may score 9 if run-off potential is neglible.	Width of riparian zone 12-18 meters throughout reach. Score high if areas < 18 meters are small or are minimally disturbed.	Width of riparian zone 6-11 meters throughout reach. Score high if areas less than 12 meters are small or are minimally disturbed.	Width of riparian zone <6 meters. Score high if areas less than 6 meters are small or are minimally disturbed.
Comments				
SCORE____(LB)	Left Bank 10 9	8 7 6	5 4 3	2 1 0
SCORE____(RB)	Right Bank 10 9	8 7 6	5 4 3	2 1 0

TOTAL SCORE _____ **Comparison to Ecoregional Guidelines** _____

If Score low, result of: _____**Natural Conditions or** _____ **Human Disturbance**

Rapid Periphyton Survey Data Sheet (Front)

Station ID: _____ **Date:** _____
Sample Log #: _____ **Sampler:** _____

Transect Number	Point	Moss	Macro	Micro	Substrate >2cm (Y/N)
1	1				
1	2				
1	3				
1	4				
1	5*				
1	6				
1	7				
1	8				
1	9				
1	10				
2	1				
2	2				
2	3				
2	4				
2	5*				
2	6				
2	7				
2	8				
2	9				
2	10				
3	1				
3	2				
3	3				
3	4				
3	5*				

Transect Number	Point	Moss	Macro	Micro	Substrate >2cm (Y/N)
3	6				
3	7				
3	8				
3	9				
3	10				
4	1				
4	2				
4	3				
4	4				
4	5*				
4	6				
4	7				
4	8				
4	9				
4	10				
5	1				
5	2				
5	3				
5	4				
5	5*				
5	6				
5	7				
5	8				
5	9				
5	10				

Canopy Cover	Trans 1	Trans 2	Trans 3	Trans 4	Trans 5
u/s					
d/s					
rdb					
ldb					
Percent (Tot/384)					

Comments:

Coverage Class (Moss and Algae)					
0	**1**	**2**	**3**	**4**	**5**
0%	<5%	5 to 25%	26 to 50%	51% to 75%	>75%

Biofilm Thickness					
0	**1**	**2**	**3**	**4**	**5**
0 mm	<0.5 mm	0.5 to 1 mm	1 to 5 mm	5 to 20 mm	> 20 mm
rough	*slimy, no visible biofilm*	*biofilm is visible*			

Substrate Size

Record "Y" in column if predominant substrate is greater than 2 cm in size.

Record "N" if not greater than 2 cm.

* Measure canopy cover at mid-point of transect

HABITATS SAMPLED (specify number of aliquots)

Riffle Rocks _____ Pool Rocks _____ Leaf Packs _____ Aquatic Plants or Roots _____
Woody Debris _____ Sediment Dep Area _____ Sand Dep Area _____ Other (specify) _____

Rapid Periphyton Survey Data Sheet (Back)

Not Necessary if Stream Survey Form has been completed

STATION NUMBER:	_____	**ASSESSORS:**	_____
STREAM NAME:	_____	**DATE:**	_____
STATION LOCATION:	_____	**TIME:**	_____
COUNTY:	_____	**STREAM MILE:**	_____
WBID#/HUC:	_____	**STREAM ORDER:**	_____
WATERSHED GROUP #	_____	**DRAINAGE AREA:**	_____
LATITUDE DEC/DEG:	_____	**GAZETTEER PAGE:**	_____
LONGITUDE DEC/DEG :	_____	**USGS QUAD:**	_____
ECOREGION:	_____	**SAMPLE LOG #**	_____
PROJECT/PURPOSE:	_____		

FIELD MEASUREMENTS

METERS USED: _____

pH	SU	DISSOLVED OXYGEN	PPM
CONDUCTIVITY	UMHOS	TIME	
TEMPERATURE	^{o}C	OTHERS	

Previous 48 hours Precip:	UNKNOWN	NONE	LITTLE	MODERATE	HEAVY	FLOODING
Ambient Weather:	SUNNY	CLOUDY	BREEZY	RAIN	SNOW	AIR TEMP:

SEDIMENT DEPOSITS:		NONE	SLIGHT	MODERATE	EXCESSIVE	BLANKET	
TYPE:	SLUDGE	MUD	SAND	SILT	NONE	OTHER_____	Contaminated Y or N
TURBIDITY:	CLEAR	SLIGHT	MODERATE	HIGH	OPAQUE		

STREAM SKETCH (include flow direction, reach distance, distance from bridge, sampling points, tribs, outfalls, livestock access, riparian area etc.)

COMMENTS:

STATE OF NESSEE - ENVIRONMENTAL LABORATORIES

Biological Analysis

Please Print Legibly

****Schedule must be arranged in advance for all tests (615) 262-6327**

Project/Site No.		**Screening Bioassays**	**Chronic Bioassays**	Branch Lab Number
Project Name		(Cannot be used for permitting)	Chronic Cd	**Chain of Custody (sign full name)**
Station No.	County	48 hr Static Screening Cd	Log Number	1. Collected by
Description		Log Number	LC50 @ 24 hrs	Date Time
Stream Mile	Depth	LC50 @ 24 hrs	LC50 @ 48 hrs	Delivered to
Collection Date	Time	LC50 @ 48 hrs	LC50 @ 72 hrs	Date Time
Sampler's name (Print)		48 hr Static Screening Pp	LC50 @ 96 hrs	2. Received by
Sampling Agency		Log Number	Survival	Date Time
Billing Code		LC50 @ 24 hrs	NOAEC	Delivered to
If Priority, Date Needed		LC50 @ 48 hrs	LOAEC	Date Time
Send Report to			Reproduction	3. Received by
		Acute Bioassays	NOAEC	Date Time
		48 hr Static Definitive Cd	LOAEC	Delivered to
Contact Hazard		Log Number	IC25	Date Time
Date Reported	By	LC50 @ 24 hrs	Chronic Pp	4. Rec'd in Lab by
Reviewed By		LC50 @ 48 hrs	Log Number	Date Time
Reviewed by		NOAEC	LC50 @ 24 hrs	Logged in by
BIOLOGICAL SURVEYS		LOAEC	LC50 @ 48 hrs	Date Time
Macroinvertebrate Recon		48 hr Static Definitive Pp	LC50 @ 72 hrs	**Additional Information**
Rapid Bioassessment (State SOP)		Log Number	LC50 @ 96 hrs	1. Approx. volume of sample
Intensive Survey - Surber		LC50 @ 24 hrs	LC50 @ 120 hrs	2. Nearest town or city
Intensive Survey - Dendy		LC50 @ 48 hrs	LC50 @ 144 hrs	
Fish Population Recon		NOAEC	LC50 @ 168 hrs	3. Others present at collection
Fish Population Intensive		LOAEC	Survival	
Fish Tissue Collection		96 hr Static Definitive Cd	NOAEC	4. Number of other samples collected at same
Chlorophyll Analysis		Log Number	LOAEC	time at this point
Log Number		LC50 @ 24 hrs	Growth	
Chlorophyll a		LC50 @ 48 hrs	NOAEC	5. Field collection procedure, handling and/or
Pheophyton		LC50 @ 72 hrs	LOAEC	preservation of this sample
SPECIAL STUDIES		LC50 @ 96 hrs	IC25	
(Please Specify)		NOAEC		
		LOAEC	Chlorine Residual	6. Mode of transportation to lab
		96 hr Static Definitive Pp		
		Log Number	Lab Parameters	7. Sample/cooler sealed by
		LC50 @ 24 hrs	pH	
		LC50 @ 48 hrs	Cond.	8. Date sample/cooler sealed
		LC50 @ 72 hrs	D.O.	9. Remarks
		LC50 @ 96 hrs	Temp.	
		NOAEC		
		LOAEC		

PH-3010 (rev 1/96)

RDA 1527

NON-DIATOM ALGAE TAXONOMIC BENCH SHEET

STATION ID:	**LAB LOG #:**
SOURCE:	**COL. BY:**
LOCATION:	**DATE COL:**
SAMPLE TYPE:	**TIME:**
ECOREGION:	**TAXONOMIST:**
SAMPLE LOG #:	**DATE:**

Phylum	Class	Order	Family	Genus, Species, Variety	Count	Biovolume

DIATOM ALGAE TAXONOMIC BENCH SHEET

STATION ID:	**LAB LOG #:**
SOURCE:	**COL. BY:**
LOCATION:	**DATE COL:**
SAMPLE TYPE:	**TIME:**
ECOREGION:	**TAXONOMIST:**
SAMPLE LOG #:	**DATE:**

Phylum	Class	Order	Family	Genus, Species, Variety	Count	Biovolume

PERIPHYTON ASSESSMENT REPORT

STATION ID:		**LOG NUMBER:**
STREAM:		**ECOREGION:**
LOCATION:		**DATE:**
HUC/ADB SEGMENT:		**WATERSHED GROUP:**
STREAM ORDER:		**DRAINAGE AREA:**
SAMPLED BY:	**ID BY:**	**SCORED BY:**

MULTI-HABITAT PERIPHYTON SAMPLE

SAMPLE TYPE (circle one): **STANDARD** (scrape) or **ALTERNATIVE** (syringe)

KDBI Metrics

METRIC	VALUE	CALCULATED SCORE
TNDT		(TNDT/102 * 100) =
H'		(H'/1.43 *100) =
KPTI		(KPTI/3.46 *100) =
CGR		(CGR/13 *100) =
FGR		(FGR/8 * 100) =
%NNS		(%NNS -%NNS)/(100-2.26)*100 =

FINAL KDBI SCORE = _____

Bioassessment Guidelines for KDBI Scores

	MT (Ecoregions 66, 67, 68, and 69)	**MVIR** (Ecoregions 73, 74, 65a, 65b, 65e, and 65i)	**PN** (Ecoregions 71 and 65j)
Excellent	75-100	57-100	67-100
Good	62-74.99	48-56.99	55-66.99
Fair	51-61.99	42-47.99	50-54.99
Poor	0-50.99	0-41.99	0-49.99

KDBI BIOASSESSMENT (circle one): **Excellent** **Good** **Fair** **Poor**

NON-DIATOM TAXA RICHNESS = _____ **TOTAL NUMBER NON-DIATOM INDIVIDUALS** = _____

HABITAT ASSESSMENT SCORE _____RR/HIGH GRAD. (or) _____GP/LOW GRAD.

HABITAT GUIDELINES FOR SUBREGION (circle one): ABOVE BELOW

COMMENTS:

APPENDIX C

PERIPHYTON METRICS

DIATOM METRICS
NON-DIATOM "SOFT ALGAE" METRICS
RAPID PERIPHYTON SURVEY METRICS

DIATOM METRICS

Source	Metric	Description	Response to increasing Impairment
Bahls 1993	# *Navicula* + *Nitzschia* spp. Counted	number of species in the genera *Navicula* and *Nitzschia*	+
Bahls 1993	Number of diatom taxa	Number of diatom taxa	-
Bahls 1993	relative abundance of dominant diatom taxon	relative abundance of individuals of the most common diatom taxon	+
Bahls 1993	relative abundance of *Navicula* + *Nitzschia* spp.	percentage of species that belong to the genera *Navicula* and *Nitzschia*	+
Fore and Grafe 2002	% *Achnanthes minutissimum*	percentage of individuals that are *Achnanthes minutissimum*	+
Fore and Grafe 2002	% alkaliphilic	See Fore and Grafe 2002	Increase with increasing pH
Fore and Grafe 2002	% dominance of top 1-5 dominant taxa	relative abundance of individuals in the 5 most common taxa	+
Fore and Grafe 2002	% eutrophic (just doing Van Dam categories 5, 6 & 7)	See Fore and Grafe 2002	+
Fore and Grafe 2002	% nitrogen fixers (only diatoms)	See Fore and Grafe 2002	-
Fore and Grafe 2002	% nitrogen heterotrophs	See Fore and Grafe 2002	-
Fore and Grafe 2002	% oligosaprobic (Van Dam category 1)	See Fore and Grafe 2002	-
Fore and Grafe 2002	% oligotrophic (Van Dam categories 1 & 2)	See Fore and Grafe 2002	-
Fore and Grafe 2002	% polysaprobic (Van Dam category 5)	See Fore and Grafe 2002	+
Fore and Grafe 2002	% require high oxygen (Van Dam categories 1 & 2)	See Fore and Grafe 2002	-
Fore and Grafe 2002	% tolerate low oxygen (Van Dam categories 4 & 5)	See Fore and Grafe 2002	+
Fore and Grafe 2002	% very + moderately motile (*Gyrosigma*, *Cylindrotheca* and others)	percentage of individuals that belong to the general *Gyrosigma*, *Cylindrotheca*, *Navicula*, *Nitzschia* and *Surirella*	+

Source	**Metric**	**Description**	**Response to increasing Impairment**
Fore and Grafe 2002	Salinity tolerance (Van Dam categories 3 & 4)	See Fore and Grafe 2002	-
Greenwood, pers. comm.	% *Eunotia* individuals	percentage of individuals that belong to *Eunotia*	
Greenwood, pers. comm.	% *Eunotia* species	percentage of the species identified in each sample that belong to *Eunotia*	
Greenwood, pers. comm.	% *Nitzschia* individuals	percentage of individuals that belong to *Nitzschia*	+
Greenwood, pers. comm.	% *Nitzschia* species	percentage of the species identified in each sample that belong to *Nitzschia*	+
Hill et al 2000	% acidophilic diatoms (1-% acidophilic diatoms)	See Hill et al 2000	Increase with decreasing pH
Hill et al 2000	% eutraphentic diatoms (1-% eutraphentic diatoms)	See Hill et al 2000	+
Hill et al 2000	Hill Dominant Diatom metric (1 - % relative abundance of dominant diatom taxon)	See Hill et al 2000	+
Hill et al 2000	Hill Motile diatoms metric (1 - % motile diatoms)	See Hill et al 2000	+
Wang et al 2005	# KY 0 spp	Number of species present belonging to KY tolerance category 0	-
Wang et al 2005	# KY 1 spp	Number of species present belonging to KY tolerance category 1	-
Wang et al 2005	# KY 2 spp	Number of species present belonging to KY tolerance category 2	-
Wang et al 2005	# KY 3 spp	Number of species present belonging to KY tolerance category 3	+

Source	**Metric**	**Description**	**Response to increasing Impairment**
Wang et al 2005	# KY 4 spp	Number of species present belonging to KY tolerance category 4	+
Wang et al 2005	# MT 1 spp	Number of species present belonging to MT tolerance category 1	+
Wang et al 2005	# MT 2 spp	Number of species present belonging to MT tolerance category 2	V
Wang et al 2005	# MT 3 spp	Number of species present belonging to MT tolerance category 3	-
Wang et al 2005	% *Achnanthes* / (*Achnanthes* + *Navicula*)	percentage of individuals in the genus *Achnanthes* divided by the percentage of individuals in the genus *Achnanthes* plus the percentage of individuals in the genus *Navicula*	-
Wang et al 2005	% *Achnanthes* individuals	percentage of individuals that belong to *Achnanthes*	-
Wang et al 2005	% *Achnanthes* species	percentage of the species identified in each sample that belong to *Achnanthes*	-
Wang et al 2005	% *Amphora* individuals	percentage of individuals that belong to *Amphora*	V
Wang et al 2005	% *Amphora* species	percentage of the species identified in each sample that belong to *Amphora*	V
Wang et al 2005	% *Cocconeis* individuals	percentage of individuals that belong to *Cocconeis*	-
Wang et al 2005	% *Cocconeis* species	percentage of the species identified in each sample that belong to *Cocconeis*	-
Wang et al 2005	% *Cyclotella* individuals	percentage of individuals that belong to *Cyclotella*	+
Wang et al 2005	% *Cyclotella* species	percentage of the species identified in each sample that belong to *Cyclotella*	-

Source	**Metric**	**Description**	**Response to increasing Impairment**
Wang et al 2005	% *Cymbella* / (*Cymbella* + *Navicula*)	percentage of individuals in the genus *Cymbella* divided by the percentage of individuals in the genus *Cymbella* plus the percentage of individuals in the genus *Navicula*	-
Wang et al 2005	% *Cymbella* individuals	percentage of individuals that belong to *Cymbella*	-
Wang et al 2005	% *Cymbella* species	percentage of the species identified in each sample that belong to *Cymbella*	-
Wang et al 2005	% *Fragilaria* individuals	percentage of individuals that belong to *Fragilaria*	-
Wang et al 2005	% *Fragilaria* species	percentage of the species identified in each sample that belong to *Fragilaria*	-
Wang et al 2005	% *Frustulia* individuals	percentage of individuals that belong to *Frustulia*	-
Wang et al 2005	% *Frustulia* species	percentage of the species identified in each sample that belong to *Frustulia*	-
Wang et al 2005	% *Gomphonema* individuals	percentage of individuals that belong to *Gomphonema*	-
Wang et al 2005	% *Gomphonema* species	percentage of the species identified in each sample that belong to *Gomphonema*	-
Wang et al 2005	% *Navicula* individuals	percentage of individuals that belong to *Navicula*	V
Wang et al 2005	% *Navicula* species	percentage of the species identified in each sample that belong to *Navicula*	V
Wang et al 2005	% *Rhoicosphenia* individuals	percentage of individuals that belong to *Rhoicosphenia*	-
Wang et al 2005	% *Rhoicosphenia* species	percentage of the species identified in each sample that belong to *Rhoicosphenia*	-
Wang et al 2005	% *Surirella* individuals	percentage of individuals that belong to *Surirella*	-
Wang et al 2005	% *Surirella* species	percentage of the species identified in each sample that belong to *Surirella*	-

Source	**Metric**	**Description**	**Response to increasing Impairment**
Wang et al 2005	% *Synedra* individuals	percentage of individuals that belong to *Synedra*	-
Wang et al 2005	% *Synedra* species	percentage of the species identified in each sample that belong to *Synedra*	-
Wang et al 2005	Evenness index	Evenness index	-
Wang et al 2005	KY 0 % of individuals	Percentage of individuals belonging to KY tolerance category 0	+
Wang et al 2005	KY 0 % of spp	Percentage of species present belonging to KY tolerance category 2	+
Wang et al 2005	KY 1 % of individuals	Percentage of individuals belonging to KY tolerance category 1	+
Wang et al 2005	KY 1 % of spp	Percentage of species present belonging to KY tolerance category 1	+
Wang et al 2005	KY 2 % of individuals	Percentage of individuals belonging to KY tolerance category 2	+
Wang et al 2005	KY 2 % of spp	Percentage of species present belonging to KY tolerance category 2	+
Wang et al 2005	KY 3 % of individuals	Percentage of individuals belonging to KY tolerance category 3	-
Wang et al 2005	KY 3 % of spp	Percentage of species present belonging to KY tolerance category 3	-
Wang et al 2005	KY 4 % of individuals	Percentage of individuals belonging to KY tolerance category 4	-
Wang et al 2005	KY 4 % of spp	Percentage of species present belonging to KY tolerance category 4	-
Wang et al 2005	MT 1 % of individuals	Percentage of individuals belonging to MT tolerance category 1	+
Wang et al 2005	MT 1 % of spp	Percentage of species present belonging to MT tolerance category 1	+
Wang et al 2005	MT 2 % of individuals	Percentage of individuals belonging to MT tolerance category 2	-
Wang et al 2005	MT 3 % of individuals	Percentage of individuals belonging to MT tolerance category 3	-

Source	**Metric**	**Description**	**Response to increasing Impairment**
Wang et al 2005	Shannon diversity index	Shannon diversity index	-
Wang et al 2005; Fore and Grafe 2002	% erect	percentage of diatom individuals with an erect growth form	-
Wang et al 2005; Fore and Grafe 2002	% prostrate	percentage of diatom individuals with a prostrate growth form	+
Wang et al 2005; Fore and Grafe 2002	% stalked	percentage of diatom individuals with a stalked growth form	V
Wang et al 2005; Fore and Grafe 2002	% unattached	percentage of diatom individuals with an unattached growth form	+
Wang et al 2005; Fore and Grafe 2002	% variable	percentage of diatom individuals with a variable growth form	V
Arnwine, pers. comm.	% Scrapers	Percent of individuals in accompanying macroinvertebrate sample that are scrapers	V

NON-DIATOM "SOFT ALGAE" METRICS

Source	Metric	Description	Response to increasing Impairment
Bahls 1993	# non-diatom divisions	Number of non-diatom divisions	-
Bahls 1993	Number of non-diatom genera	Number of non-diatom genera	-
Bahls 1993	Dominant phylum	Non-diatom phylum with greatest abundance in sample	V
Greenwood, 2008	% nitrogen fixers (all algae)	See Fore and Grafe 2002	-
Hill et al 2000	% diatoms	See Hill et al 2000	-
Hill et al 2000	Hill Cyanobacteria metric (1 - % cyanobacteria)	See Hill et al 2000	+
Hill et al 2000	Hill PIBI	See Hill et al 2000	-
Hill et al 2000	Relative taxon richness	Relative taxon richness	+
Arnwine, pers. Comm.	% Scrapers	Percent of individuals in accompanying macroinvertebrate sample that are scrapers	V

RAPID PERIPHYTON SURVEY METRICS

Source	Metric	Description	Response to increasing Impairment
USEPA, 1999	Mean Moss Coverage Class	Mean of moss coverage class scores	+
USEPA, 1999	Mean Macroalgae Coverage Class	Mean of all macroalgae coverage class scores	+
USEPA, 1999	Mean Microalgae Thickness	Mean of all microalgae thickness scores	+
USEPA, 1999	% Nutrient Tolerant	% of macroinvertebrates from accompanying sample that are Nutrient Tolerant	+
USEPA, 1999	% Scrapers	Percent of individuals in accompanying macroinvertebrate sample that are scrapers	+
USEPA, 1999	% Cover by Moss	% of points that have moss coverage	+
USEPA, 1999	% Cover by Macroalgae	% of points that have macroalgae coverage	+
USEPA, 1999	% Cover by Microalgae	% of points over suitable substrate that have microalgae coverage	+

APPENDIX D

TAXONOMIC INFORMATION

TAXONOMIC KEYS
TAXONOMIC SPECIALISTS FOR REFERENCE VERIFICATION
VERIFIED TAXA LIST FOR DIATOMS -
(INCLUDING KENTUCKY POLLUTION TOLERANCE INDEX VALUES)
VERIFIED TAXA LIST FOR SOFT ALGAE (NON-DIATONS)

TAXONOMIC KEYS

Camburn, K.E., R.L. Lowe, and D.L. Stoneburner. 1978. The haptobenthic diatom flora of Long Branch Creek, South Carolina. *Nova Hedwigia* 30:149-279.

Collins, G.B. and R.G. Kalinsky. 1977. Studies on Ohio diatoms: I. Diatoms of the Scioto River Basin. *Bull. Ohio Biological Survey*. 5(3):1-45.

Cox, E. J. 1996. *Identification of freshwater diatoms from live material*. Chapman & Hall, London.

Czarnecki, D.B. and D.W. Blinn. 1978. *Diatoms of the Colorado River in Grand Canyon National Park and vicinity*. (Diatoms of Southwestern USA II). Bibliotheca Phycologia 38. J. Cramer. 181 pp.

Dawes, C. J. 1974. *Marine Algae of the West Coast of Florida*. University of Miami Press.

Dillard, G.E. 1989a. Freshwater algae of the Southeastern United States. Part 1. Chlorophyceae: Volvocales, Testrasporales, and Chlorococcales. *Bibliotheca*, 81.

Dillard, G.E. 1989b. Freshwater algae of the Southeastern United States. Part 2. Chlorophyceae: Ulotrichales, Microsporales, Cylindrocapsales, Sphaeropleales, Chaetophorales, Cladophorales, Schizogoniales, Siphonales, and Oedogoniales. *Bibliotheca Phycologica*, 83.

Dillard, G.E. 1990. Freshwater algae of the Southeastern United States. Part 3. Chlorophyceae: Zygnematales: Zygenmataceae, Mesotaeniaceae, and Desmidaceae (Section 1). *Bibliotheca Phycologica*, 85.

Dillard, G.E. 1991. Freshwater algae of the Southeastern United States. Part 4. Chlorophyceae: Zygnemateles: Desmidaceae (Section 2). *Bibliotheca Phycologica*, 89.

Drouet, F. 1968. *Revision of the classification of the oscillatoriaceae*. Monograph 15. Academy of Natural Sciences, Philadelphia. Fulton Press, Lancaster, Pennsylvania.

Hohn, M.H. and J. Hellerman. 1963. The taxonomy and structure of diatom populations from three North American rivers using three sampling methods. *Transaction of the American Microscopal Society* 82:250-329.

Hustedt, F. 1927-1966. Die kieselalgen In Rabenhorst's Kryptogamen-flora von Deutschland Osterreich und der Schweiz VII. Leipzig, West Germany.

TAXONOMIC KEYS (cont.)

Hustedt, F. 1930. *Bacillariophyta (Diatomae)*. In Pascher, A. (ed). Die suswasser Flora Mitteleuropas. (The freshwater flora of middle Europe). Gustav Fischer Verlag, Jena, Germany.

Jarrett, G.L. and J.M. King. 1989. The diatom flora (Bacillariphyceae) of Lake Barkley. U.S. Army
Corps of Engineers, Nashville Dist. #DACW62-84-C-0085.

Krammer, K. and H. Lange-Bertalot. 1986-1991. Susswasserflora von Mitteleuropa. Band 2. Parts
1-4. Bacillariophyceae. Gustav Fischer Verlag. Stuttgart. New York.

Lange-Bertalot, H. and R. Simonsen. 1978. A taxonomic revision of the Nitzschia lanceolatae Grunow: 2. European and related extra-European freshwater and brackish water taxa. *Bacillaria* 1:11-111.

Lange-Bertalot, H. 1980. New species, combinations and synonyms in the genus Nitzschia. *Bacillaria* 3:41-77.

Patrick, R. and C.W. Reimer. 1966. *The diatoms of the United States, exclusive of Alaska and Hawaii*. Monograph No. 13. Academy of Natural Sciences, Philadelphia, Pennsylvania.

Patrick, R. and C.W. Reimer. 1975. *The Diatoms of the United States*. Vol. 2, Part 1. Monograph No. 13. Academy of Natural Sciences, Philadelphia, Pennsylvania.

Prescott, G.W. 1962. *The algae of the Western Great Lakes area*. Wm. C. Brown Co., Dubuque, Iowa.

Prescott, G.W., H.T. Croasdale, and W.C. Vinyard. 1975. *A Synopsis of North American desmids. Part II. Desmidaceae: Placodermae*. Section 1. Univ. Nebraska Press, Lincoln, Nebraska.

Prescott, G.W., H.T. Croasdale, and W.C. Vinyard. 1977. *A synopsis of North American desmids. Part II. Desmidaceae: Placodermae*. Section 2. Univ. Nebraska Press, Lincoln, Nebraska.

Prescott, G.W., H.T. Croasdale, and W.C. Vinyard. 1981. *A synopsis of North American desmids. Part II. Desmidaceae: Placodermae*. Section 3. Univ. Nebraska Press, Lincoln, Nebraska.

Prescott, G.W. 1978. *How to know the freshwater algae*. 3rd Edition. Wm. C. Brown Co., Dubuque, Iowa.

Simonsen, R. 1987. *Atlas and catalogue of the diatom types of Friedrich Hustedt*. Vol. 1-3. J. Cramer. Berlin, Germany.

TAXONOMIC KEYS (cont.)

Smith, M. 1950. *The Freshwater Algae of the United States*. McGraw-Hill, New York, New York.

Taylor, W. R. 1960. *Marine algae of the eastern tropical and subtropical coasts of the Americas*. University of Michigan Press, Ann Arbor, Michigan.

VanLandingham, S. L. 1982. *Guide to the identification, environmental requirements and pollution tolerance of freshwater blue-green algae (Cyanophyta)*. EPA-600/3-82-073.

Whitford, L.A. and G.J. Schumacher. 1973. *A manual of freshwater algae*. Sparks Press, Raleigh, North Carolina.

Wujek, D.E. and R.F. Rupp. 1980. Diatoms of the Tittabawassee River, Michigan. *Bibliotheca Phycologia* 50:1-100.

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DIATOM TAXA LIST (Sorted by Genus)

Order	Family	Genus	Species	KPTI Value
Acnanthales	Achnanthaceae	Achnanthes	childanos	0
Acnanthales	Achnanthaceae	Achnanthes	conspicua	3
Acnanthales	Achnanthaceae	Achnanthes	deflexa	4
Acnanthales	Achnanthaceae	Achnanthes	deflexa var. alpestris	4
Acnanthales	Achnanthaceae	Achnanthes	exigua	4
Acnanthales	Achnanthaceae	Achnanthes	hustedtii	4
Acnanthales	Achnanthaceae	Achnanthes	inflata	0
Acnanthales	Achnanthaceae	Achnanthes	laevis	3
Pennales	Achnanthaceae	Achnanthes	minutissima var. scotica	
Acnanthales	Achnanthaceae	Achnanthes	ricula	0
Pennales	Achnanthaceae	Achnanthes	rupestoides	
Acnanthales	Achnanthaceae	Achnanthes	solea	0
Acnanthales	Achnanthaceae	Achnanthes	sp.	3
Acnanthales	Achnanthaceae	Achnanthes	stewartii	4
Acnanthales	Achnanthaceae	Achnanthes	subatomoides	0
Pennales	Achnanthaceae	Achnanthes	subhudsonis	
Acnanthales	Achnanthaceae	Achnanthes	subhudsonis var. kraeuselii	3
Pennales	Achnanthidiaceae	Achnanthidium	affine	
Pennales	Achnanthidiaceae	Achnanthidium	altergracillima	
Pennales	Achnanthidiaceae	Achnanthidium	caledonicum	
Pennales	Achnanthidiaceae	Achnanthidium	catenatum	
Acnanthales	Achnanthidiaceae	Achnanthidium	coarctatum	0
Pennales	Achnanthidiaceae	Achnanthidium	deflexum	
Acnanthales	Achnanthidiaceae	Achnanthidium	delicatulum	0
Pennales	Achnanthidiaceae	Achnanthidium	exiguum	
Pennales	Achnanthidiaceae	Achnanthidium	exilis	
Pennales	Achnanthidiaceae	Achnanthidium	latecephalum	
Acnanthales	Achnanthidiaceae	Achnanthidium	macrocephalum	3
Acnanthales	Achnanthidiaceae	Achnanthidium	minutissimum	3
Pennales	Achnanthidiaceae	Achnanthidium	rivulare	
Coscinodiscales	Hemidiscaceae	Actinocyclus	normanii	0
Pennales	Naviculaceae	Adlafia	bryophila	
Pennales	Naviculaceae	Adlafia	minuscula	
Pennales	Naviculaceae	Adlafia	minuscula var. muralis	
Pennales	Naviculaceae	Adlafia	multnomahii	
Pennales	Naviculaceae	Adlafia	suchlandtii	
Naviculales	Amphipleuraceae	Amphipleura	pellucida	3

Order	Family	Genus	Species	KPTI Value
Thalassiophysales	Catenulaceae	Amphora	bullatoides	0
Pennales	Catenulaceae	Amphora	copulata	
Thalassiophysales	Catenulaceae	Amphora	libyca	3
Thalassiophysales	Catenulaceae	Amphora	montana	3
Thalassiophysales	Catenulaceae	Amphora	ovalis	3
Thalassiophysales	Catenulaceae	Amphora	pediculus	3
Thalassiophysales	Catenulaceae	Amphora	sp.	3
Thalassiophysales	Catenulaceae	Amphora	veneta	1
Mastogloiales	Mastogloiaceae	Aneumastus	tusculus	0
Pennales	Fragilariaceae	Asterionella	formosa	3
Centrales	Aulacoseiraceae	Aulacoseira	alpigena	3
Centrales	Aulacoseriaceae	Aulacoseira	ambigua	
Centrales	Aulacoseriaceae	Aulacoseira	crenulata	
Centrales	Aulacoseiraceae	Aulacoseira	distans	3
Centrales	Aulacoseiraceae	Aulacoseira	granulata	3
Centrales	Aulacoseiraceae	Aulacoseira	granulata var. angustissima	3
Centrales	Aulacoseiraceae	Aulacoseira	italica	3
Centrales	Aulacoseriaceae	Aulacoseira	subborealis	
Bacillariales	Bacillariaceae	Bacillaria	paradoxa	2
Pennales	Brachysiraceae	Brachysira	apiculata	
Naviculales	Brachysiraceae	Brachysira	serians	0
Naviculales	Brachysiraceae	Brachysira	vitrea	2
Pennales	Pinnulariaceae	Caloneis	bacillum	3
Pennales	Pinnulariaceae	Caloneis	schumanniana	
Pennales	Pinnulariaceae	Caloneis	silicula	
Surirellales	Surirellaceae	Campylodiscus	hibernicus	0
Naviculales	Naviculaceae	Capartogramma	crucicula	2
Naviculales	Cavinulaceae	Cavinula	cocconeiformis	0
Pennales	Cavinulaceae	Cavinula	jaernefelti	
Naviculales	Cavinulaceae	Cavinula	lacustris	0
Pennales	Naviculaceae	Chamaepinnularia	mediocris	
Pennales	Achanthaceae	Cocconeis	neodiminuta	
Acnanthales	Cocconeidaceae	Cocconeis	pediculus	3
Pennales	Achanthaceae	Cocconeis	placentula	3
Acnanthales	Cocconeidaceae	Cocconeis	placentula var. euglypta	3
Acnanthales	Cocconeidaceae	Cocconeis	placentula var. lineata	3
Acnanthales	Cocconeidaceae	Cocconeis	placentula var. pseudolineata	3
Naviculales	Stauroneidaceae	Craticula	accomoda	1
Pennales	Stauroneidaceae	Craticula	ambigua	

Order	Family	Genus	Species	KPTI Value
Pennales	Stauroneidaceae	Craticula	citrus	
Naviculales	Stauroneidaceae	Craticula	cuspidata	2
Naviculales	Stauroneidaceae	Craticula	halophila	2
Pennales	Stauroneidaceae	Craticula	halophiliodes	
Pennales	Stauroneidaceae	Craticula	minuscloides	
Pennales	Stauroneidaceae	Craticula	molestiformis	
Naviculales	Stauroneidaceae	Craticula	submolesta	1
Pennales	Fragilariaceae	Ctenophora	pulchella	1
Pennales	Fragilariaceae	Ctenophora	pulchella var. lacerata	1
Centrales	Stephanodiscaceae	Cyclostephanos	dubius	3
Centrales	Stephanodiscaceae	Cyclostephanos	invisitatus	3
Centrales	Stephanodiscaceae	Cyclostephanos	tholiformis	
Centrales	Stephanodiscaceae	Cyclotella	atomus	2
Centrales	Stephanodiscaceae	Cyclotella	meneghiniana	1
Centrales	Stephanodiscaceae	Cyclotella	ocellata	
Centrales	Stephanodiscaceae	Cyclotella	pseudostelligera	2
Centrales	Stephanodiscaceae	Cyclotella	sp.	2
Centrales	Stephanodiscaceae	Cyclotella	stelligera	3
Centrales	Stephanodiscaceae	Cyclotella	striata	4
Centrales	Stephanodiscaceae	Cyclotella	striata var. ambigua	2
Bacillariales	Bacillariaceae	Cylindrotheca	gracilis	3
Surirellales	Surirellaceae	Cymatopleura	elliptica	3
Surirellales	Surirellaceae	Cymatopleura	solea	3
Pennales	Surirellaceae	Cymatopleura	solea var. apiculata	
Pennales	Cymbellaceae	Cymbella	aequalis	
Cymbellales	Cymbellaceae	Cymbella	affinis	4
Cymbellales	Cymbellaceae	Cymbella	amphicephala	4
Cymbellales	Cymbellaceae	Cymbella	aspera	4
Pennales	Cymbellaceae	Cymbella	caespitosa	
Cymbellales	Cymbellaceae	Cymbella	cistula	4
Cymbellales	Cymbellaceae	Cymbella	cuspidata	4
Cymbellales	Cymbellaceae	Cymbella	cymbiformis	4
Cymbellales	Cymbellaceae	Cymbella	delicatula	4
Pennales	Cymbellaceae	Cymbella	ehrenbergii	
Cymbellales	Cymbellaceae	Cymbella	hauckii	0
Cymbellales	Cymbellaceae	Cymbella	hebridica	4
Pennales	Cymbellaceae	Cymbella	helvetica	
Cymbellales	Cymbellaceae	Cymbella	hustedtii	0
Cymbellales	Cymbellaceae	Cymbella	laevis	0

Order	Family	Genus	Species	KPTI Value
Cymbellales	Cymbellaceae	Cymbella	lanceolata	4
Pennales	Cymbellaceae	Cymbella	lata	
Cymbellales	Cymbellaceae	Cymbella	leptoceros	4
Pennales	Cymbellaceae	Cymbella	mesiana	
Pennales	Cymbellaceae	Cymbella	mexicana	0
Cymbellales	Cymbellaceae	Cymbella	microcephala	4
Pennales	Cymbellaceae	Cymbella	microcephala var. crassa	
Cymbellales	Cymbellaceae	Cymbella	naviculiformis	4
Pennales	Cymbellaceae	Cymbella	obscura	
Cymbellales	Cymbellaceae	Cymbella	sp.	4
Cymbellales	Cymbellaceae	Cymbella	subaequalis	0
Cymbellales	Cymbellaceae	Cymbella	subcuspidata	4
Cymbellales	Cymbellaceae	Cymbella	tumida	4
Cymbellales	Cymbellaceae	Cymbella	turgidula	4
Pennales	Not Designated	Decussata	placenta	
Bacillariales	Bacillariaceae	Denticula	elegans	3
Bacillariales	Bacillariaceae	Denticula	kuetzingii	3
Bacillariales	Bacillariaceae	Denticula	sp.	3
Pennales	Bacillariaceae	Denticula	tenuis	
Naviculales	Diadesmidaceae	Diadesmis	confervacea	2
Naviculales	Diadesmidaceae	Diadesmis	confervacea var. peregrin	2
Naviculales	Diadesmidaceae	Diadesmis	contenta	2
Naviculales	Diadesmidaceae	Diadesmis	contenta var. biceps	2
Pennales	Diadesmidaceae	Diadesmis	gallica	
Pennales	Diadesmidaceae	Diadesmis	laevissima	
Pennales	Fragilariaceae	Diatoma	hyemalis	1
Pennales	Fragilariaceae	Diatoma	moniliformis	
Pennales	Fragilariaceae	Diatoma	tenuis	0
Pennales	Fragilariaceae	Diatoma	vulgaris	3
Pennales	Cymbellaceae	Didymosphenia	geminata	
Naviculales	Diploneidaceae	Diploneis	elliptica	3
Naviculales	Diploneidaceae	Diploneis	finnica	0
Naviculales	Diploneidaceae	Diploneis	oblonella	3
Pennales	Diploneidaceae	Diploneis	parma	
Pennales	Diploneidaceae	Diploneis	peterseni	
Pennales	Diploneidaceae	Diploneis	pseudovalis	
Naviculales	Diploneidaceae	Diploneis	puella	0
Naviculales	Diploneidaceae	Diploneis	smithii var.dilatata	0
Naviculales	Diploneidaceae	Diploneis	sp.	3

Order	Family	Genus	Species	KPTI Value
Naviculales	Diploneidaceae	Diploneis	subovalis	0
Centrales	Thalassiosiraceae	Discostella	stelligera	
Pennales	Cymbellaceae	Encyonema	auerswaldii	
Pennales	Cymbellaceae	Encyonema	brehmii	
Cymbellales	Cymbellaceae	Encyonema	caespitosum	4
Cymbellales	Cymbellaceae	Encyonema	lunatum	4
Cymbellales	Cymbellaceae	Encyonema	mesianum	3
Cymbellales	Cymbellaceae	Encyonema	minutum	3
Cymbellales	Cymbellaceae	Encyonema	muelleri	4
Pennales	Cymbellaceae	Encyonema	perpussilum	
Cymbellales	Cymbellaceae	Encyonema	prostratum	4
Pennales	Cymbellaceae	Encyonema	reichardtii	
Cymbellales	Cymbellaceae	Encyonema	silesiacum	4
Cymbellales	Cymbellaceae	Encyonema	triangulum	4
Cymbellales	Cymbellaceae	Encyonemopsis	cesatii	4
Pennales	Not Designated	Encyonopsis	cesatii	
Pennales	Not Designated	Encyonopsis	falaisensis	
Pennales	Not Designated	Encyonopsis	krammeri	
Pennales	Not Designated	Encyonopsis	microcephala	
Surirellales	Entomoneidaceae	Entomoneis	alata	1
Surirellales	Entomoneidaceae	Entomoneis	ornata	1
Rhopalodiales	Rhopalodiaceae	Epithemia	adnata	2
Rhopalodiales	Rhopalodiaceae	Epithemia	adnata var. saxonica	0
Rhopalodiales	Rhopalodiaceae	Epithemia	argus	1
Rhopalodiales	Rhopalodiaceae	Epithemia	argus var. protracta	1
Pennales	Rhopalodiaceae	Epithemia	reichelti	
Rhopalodiales	Rhopalodiaceae	Epithemia	sorex	3
Rhopalodiales	Rhopalodiaceae	Epithemia	sp.	2
Rhopalodiales	Rhopalodiaceae	Epithemia	turgida	3
Rhopalodiales	Rhopalodiaceae	Epithemia	turgida var. granulata	3
Pennales	Achnanthidiaceae	Eucocconeis	flexella	
Eunotiales	Eunotiaceae	Eunotia	arcus	2
Eunotiales	Eunotiaceae	Eunotia	bilunaris	3
Pennales	Eunotiaceae	Eunotia	bilunaris var. mucophila	
Pennales	Eunotiaceae	Eunotia	carolina	
Pennales	Eunotiaceae	Eunotia	circumborealis	
Pennales	Eunotiaceae	Eunotia	denticulata	
Eunotiales	Eunotiaceae	Eunotia	exigua	2
Pennales	Eunotiaceae	Eunotia	exigua var. bidens	

Order	Family	Genus	Species	KPTI Value
Pennales	Eunotiaceae	Eunotia	fallax	
Pennales	Eunotiaceae	Eunotia	flexuosa	
Eunotiales	Eunotiaceae	Eunotia	formica	0
Pennales	Eunotiaceae	Eunotia	glacialis	
Pennales	Eunotiaceae	Eunotia	implicata	
Eunotiales	Eunotiaceae	Eunotia	incisa	0
Pennales	Eunotiaceae	Eunotia	intermedia	
Eunotiales	Eunotiaceae	Eunotia	maior	3
Pennales	Eunotiaceae	Eunotia	meisteri	
Pennales	Eunotiaceae	Eunotia	microcephala	
Pennales	Eunotiaceae	Eunotia	microcephala var. tridentata	
Pennales	Eunotiaceae	Eunotia	minor	
Pennales	Eunotiaceae	Eunotia	monodon	
Pennales	Eunotiaceae	Eunotia	monodon var. constricta	
Eunotiales	Eunotiaceae	Eunotia	muscicola	3
Eunotiales	Eunotiaceae	Eunotia	muscicola var. tridentula	3
Eunotiales	Eunotiaceae	Eunotia	naegelii	0
Pennales	Eunotiaceae	Eunotia	paludosa	
Pennales	Eunotiaceae	Eunotia	paludosa var. trinacria	
Eunotiales	Eunotiaceae	Eunotia	pectinalis	3
Eunotiales	Eunotiaceae	Eunotia	pectinalis var. minor	3
Eunotiales	Eunotiaceae	Eunotia	pectinalis var. undulata	3
Pennales	Eunotiaceae	Eunotia	perminuta	
Pennales	Eunotiaceae	Eunotia	perpusilla	3
Pennales	Eunotiaceae	Eunotia	pirla	
Eunotiales	Eunotiaceae	Eunotia	praerupta	0
Eunotiales	Eunotiaceae	Eunotia	rhomboidea	4
Eunotiales	Eunotiaceae	Eunotia	septentrionalis	0
Eunotiales	Eunotiaceae	Eunotia	serra var. diadema	3
Pennales	Eunotiaceae	Eunotia	soleirolii	
Pennales	Eunotiaceae	Eunotia	sp.	3
Pennales	Eunotiaceae	Eunotia	sudetica	
Eunotiales	Eunotiaceae	Eunotia	tenella	3
Eunotiales	Eunotiaceae	Eunotia	triodon	0
Pennales	Eunotiaceae	Eunotia	valida	
Eunotiales	Eunotiaceae	Eunotia	vanheurckii var. intermedia	0
Pennales	Eunotiaceae	Eunotia	zasuminensis	
Naviculales	Sellaphoraceae	Fallacia	auriculata	3
Naviculales	Sellaphoraceae	Fallacia	helensis	3

Order	Family	Genus	Species	KPTI Value
Pennales	Sellaphoraceae	Fallacia	lenzii	
Pennales	Sellaphoraceae	Fallacia	monoculata	
Pennales	Sellaphoraceae	Fallacia	omissa	
Naviculales	Sellaphoraceae	Fallacia	pygmaea	3
Naviculales	Sellaphoraceae	Fallacia	subhamulata	3
Naviculales	Sellaphoraceae	Fallacia	tenera	2
Naviculales	Naviculaceae	Fistulifera	pelliculosa	2
Pennales	Fragilariaceae	Fragilaria	capucina	4
Pennales	Fragilariaceae	Fragilaria	capucina var. gracile	4
Pennales	Fragilariaceae	Fragilaria	capucina var. gracilis	4
Pennales	Fragilariaceae	Fragilaria	capucina var. mesolepta	2
Pennales	Fragilariaceae	Fragilaria	capucina var. rumpens	4
Pennales	Fragilariaceae	Fragilaria	capucina var. vaucheriae	2
Pennales	Fragilariaceae	Fragilaria	capucina var. vaucheriae	4
Pennales	Fragilariaceae	Fragilaria	crotonensis	0
Pennales	Fragilariaceae	Fragilaria	delicatissima	4
Pennales	Fragilariaceae	Fragilaria	dilatata	0
Pennales	Fragilariaceae	Fragilaria	famelica	
Pennales	Fragilariaceae	Fragilaria	fasciculata	1
Pennales	Fragilariaceae	Fragilaria	nanana	3
Pennales	Fragilariaceae	Fragilaria	neoproducta	
Pennales	Fragilariaceae	Fragilaria	parasitica	4
Pennales	Fragilariaceae	Fragilaria	pseudoconstruens	
Pennales	Fragilariaceae	Fragilaria	sepes	
Pennales	Fragilariaceae	Fragilaria	sp.	3
Pennales	Fragilariaceae	Fragilaria	tenera	
Pennales	Fragilariaceae	Fragilaria	ulna	3
Pennales	Fragilariaceae	Fragilaria	vaucheriae	2
Pennales	Fragilariaceae	Fragilaria	virescens	2
Pennales	Diatomaceae	Fragilariforma	bicapitata	
Pennales	Diatomaceae	Fragilariforma	virescens	
Pennales	Amphipleuraceae	Frustulia	amphipleuroides	
Naviculales	Amphipleuraceae	Frustulia	assymetrica	0
Pennales	Amphipleuraceae	Frustulia	crassinervia	
Pennales	Amphipleuraceae	Frustulia	krammeri	
Naviculales	Amphipleuraceae	Frustulia	rhomboides	3
Naviculales	Amphipleuraceae	Frustulia	rhomboides var. amphipleuroides	3
Naviculales	Amphipleuraceae	Frustulia	rhomboides var. capitata	3

Order	Family	Genus	Species	KPTI Value
Naviculales	Amphipleuraceae	Frustulia	rhomboides var. crassinervia	4
Naviculales	Amphipleuraceae	Frustulia	rhomboides var. saxonica	3
Naviculales	Amphipleuraceae	Frustulia	saxonica	3
Naviculales	Amphipleuraceae	Frustulia	sp.	3
Naviculales	Amphipleuraceae	Frustulia	vulgaris	3
Naviculales	Amphipleuraceae	Frustulia	weinholdii	3
Pennales	Naviculaceae	Geissleria	aikenensis	
Pennales	Naviculaceae	Geissleria	decussis	
Pennales	Naviculaceae	Geissleria	schoenfeldii	
Pennales	Gomphonemataceae	Gomphoneis	herculeana	0
Pennales	Gomphonemataceae	Gomphoneis	herculeana var. loweii	
Pennales	Gomphonemataceae	Gomphoneis	herculeana var. robusta	0
Cymbellales	Gomophonemataceae	Gomphoneis	minutum	0
Cymbellales	Gomophonemataceae	Gomphonema	acuminatum	4
Cymbellales	Gomophonemataceae	Gomphonema	acuminatum var. coronatum	4
Pennales	Gomphonemataceae	Gomphonema	acuminatum var. coronatum	4
Cymbellales	Gomophonemataceae	Gomphonema	acuminatum var. elongatum	4
Cymbellales	Gomophonemataceae	Gomphonema	affine	3
Cymbellales	Gomophonemataceae	Gomphonema	angustatum	2
Cymbellales	Gomophonemataceae	Gomphonema	angustatum var. productum	2
Cymbellales	Gomophonemataceae	Gomphonema	angustum	1
Cymbellales	Gomophonemataceae	Gomphonema	apuncto	2
Cymbellales	Gomophonemataceae	Gomphonema	augur	2
Cymbellales	Gomophonemataceae	Gomphonema	clavatum	2
Cymbellales	Gomophonemataceae	Gomphonema	clavatum var. mexicanum	3
Cymbellales	Gomophonemataceae	Gomphonema	clevei	3
Pennales	Gomphonemataceae	Gomphonema	dichotomum	1
Cymbellales	Gomophonemataceae	Gomphonema	drutelingense	
Pennales	Gomphonemataceae	Gomphonema	freesei	
Cymbellales	Gomophonemataceae	Gomphonema	gracile	3
Cymbellales	Gomophonemataceae	Gomphonema	grovei var. lingulatum	4
Cymbellales	Gomophonemataceae	Gomphonema	hebridense var. sphaerophorum	4
Pennales	Gomphonemataceae	Gomphonema	insigne	
Cymbellales	Gomophonemataceae	Gomphonema	instabilis	3
Cymbellales	Gomophonemataceae	Gomphonema	intricatum	3
Cymbellales	Gomophonemataceae	Gomphonema	intricatum var. pulvinatum	3
Pennales	Gomphonemataceae	Gomphonema	kobayasii	
Pennales	Gomphonemataceae	Gomphonema	lagenula	
Cymbellales	Gomophonemataceae	Gomphonema	manubrium	4

Order	Family	Genus	Species	KPTI Value
Cymbellales	Gomophonemataceae	Gomphonema	mehleri	0
Pennales	Gomphonemataceae	Gomphonema	micropus	
Cymbellales	Gomophonemataceae	Gomphonema	minutum	2
Pennales	Gomphonemataceae	Gomphonema	olivaceoides	0
Cymbellales	Gomophonemataceae	Gomphonema	olivaceum	2
Cymbellales	Gomophonemataceae	Gomphonema	olivaceum var. minutissimum	0
Pennales	Gomphonemataceae	Gomphonema	parvulius	
Cymbellales	Gomophonemataceae	Gomphonema	parvulum	1
Pennales	Gomphonemataceae	Gomphonema	patrickii	
Pennales	Gomphonemataceae	Gomphonema	productum	
Pennales	Gomphonemataceae	Gomphonema	pseudotenellum	
Cymbellales	Gomophonemataceae	Gomphonema	puiggarianum var. aequatorialis	2
Pennales	Gomphonemataceae	Gomphonema	pumilum	
Pennales	Gomphonemataceae	Gomphonema	rhombicum	4
Cymbellales	Gomophonemataceae	Gomphonema	sp.	3
Cymbellales	Gomophonemataceae	Gomphonema	sparsistriatum f. maculatum	4
Pennales	Gomphonemataceae	Gomphonema	sphaerophorum	4
Pennales	Gomphonemataceae	Gomphonema	subclavatum	2
Pennales	Gomphonemataceae	Gomphonema	subtile	
Cymbellales	Gomophonemataceae	Gomphonema	tergestinum	0
Cymbellales	Gomophonemataceae	Gomphonema	truncatum	4
Cymbellales	Gomophonemataceae	Gomphonema	truncatum var. capitatum	4
Cymbellales	Gomophonemataceae	Gomphonema	truncatum var. turgidulum	4
Pennales	Gomphonemataceae	Gomphonema	turris	
Pennales	Gomphonemataceae	Gomphonema	ventricosum	
Pennales	Not Designated	Gomphosphenia	grovei	
Pennales	Not Designated	Gomphosphenia	grovei var. lingulata	
Pennales	Not Designated	Gomphosphenia	lingulatiformis	
Pennales	Not Designated	Gomphosphenia	tackei	
Naviculales	Pleurosigmataceae	Gyrosigma	acuminatum	3
Naviculales	Pleurosigmataceae	Gyrosigma	attenuatum	3
Naviculales	Pleurosigmataceae	Gyrosigma	nodiferum	4
Naviculales	Pleurosigmataceae	Gyrosigma	obscurum	0
Naviculales	Pleurosigmataceae	Gyrosigma	obtusatum	0
Naviculales	Pleurosigmataceae	Gyrosigma	parkerii	2
Naviculales	Pleurosigmataceae	Gyrosigma	scalproides	3
Naviculales	Pleurosigmataceae	Gyrosigma	sciotense	0
Naviculales	Pleurosigmataceae	Gyrosigma	sp.	3
Naviculales	Pleurosigmataceae	Gyrosigma	spencerii	3

Order	Family	Genus	Species	KPTI Value
Naviculales	Pleurosigmataceae	Gyrosigma	spencerii var. curvula	3
Bacillariales	Bacillariaceae	Hantzschia	amphioxys	3
Pennales	Bacillariaceae	Hantzschia	distinctepunctata	
Bacillariales	Bacillariaceae	Hantzschia	elongata	0
Naviculales	Naviculaceae	Hippodonta	capitata	3
Pennales	Naviculaceae	Hippodonta	hungarica	
Naviculales	Naviculaceae	Hippodonta	luneburgensis	3
Acnanthales	Achnanthidiaceae	Karayevia	clevei	4
Acnanthales	Achnanthidiaceae	Karayevia	clevei var. rostrata	4
Pennales	Achnanthaceae	Karayevia	laterostrata	
Acnanthales	Achnanthidiaceae	Karayevia	suchlandtii	4
Acnanthales	Achnanthidiaceae	Lemnicola	hungarica	4
Naviculales	Diadesmidaceae	Luticola	cohnii	1
Pennales	Diadesmidaceae	Luticola	goeppertiana	
Naviculales	Diadesmidaceae	Luticola	mutica	2
Naviculales	Diadesmidaceae	Luticola	mutica var. binodis	2
Naviculales	Diadesmidaceae	Luticola	mutica var. stigma	2
Naviculales	Diadesmidaceae	Luticola	mutica var. undulata	2
Naviculales	Diadesmidaceae	Luticola	muticavar. ventricosa	2
Naviculales	Diadesmidaceae	Luticola	nivalis	2
Naviculales	Diadesmidaceae	Luticola	saxophila	0
Pennales	Diadesmidaceae	Luticola	ventricosa	
Mastogloiales	Mastogloiaceae	Mastogloia	smithii	0
Pennales	Naviculaceae	Mayamaea	atomus	1
Pennales	Naviculaceae	Mayamaea	atomus var. permitis	
Centrales	Melosiraceae	Melosira	varians	2
Pennales	Fragilariaceae	Meridion	circulare	3
Pennales	Fragilariaceae	Meridion	circulare var. constrictum	3
Cymbellales	Cymbellaceae	Navicella	pusilla	0
Naviculales	Naviculaceae	Navicula	aboensis	
Pennales	Naviculaceae	Navicula	absoluta	
Naviculales	Naviculaceae	Navicula	agrestis	0
Naviculales	Naviculaceae	Navicula	angusta	0
Pennales	Naviculaceae	Navicula	antonii	
Naviculales	Naviculaceae	Navicula	arvensis	3
Pennales	Naviculaceae	Navicula	cananlis	
Naviculales	Naviculaceae	Navicula	capitatoradiata	2
Naviculales	Naviculaceae	Navicula	cari	1
Naviculales	Naviculaceae	Navicula	caterva	2

Order	Family	Genus	Species	KPTI Value
Naviculales	Naviculaceae	Navicula	clementioides	0
Naviculales	Naviculaceae	Navicula	clementis	0
Pennales	Naviculaceae	Navicula	cryptocephala	3
Naviculales	Naviculaceae	Navicula	cryptocephala var.exilis	4
Naviculales	Naviculaceae	Navicula	cryptotenella	2
Naviculales	Naviculaceae	Navicula	decussis	3
Pennales	Naviculaceae	Navicula	detenta	
Naviculales	Naviculaceae	Navicula	dibola	0
Pennales	Naviculaceae	Navicula	difficillima	
Pennales	Naviculaceae	Navicula	diluviana	
Naviculales	Naviculaceae	Navicula	elginensis	3
Naviculales	Naviculaceae	Navicula	elginensis var. neglecta	3
Naviculales	Naviculaceae	Navicula	elginensis var. rostrata	3
Naviculales	Naviculaceae	Navicula	erifuga	2
Pennales	Naviculaceae	Navicula	evanida	
Naviculales	Naviculaceae	Navicula	exigua	3
Naviculales	Naviculaceae	Navicula	exigua var. capitata	3
Naviculales	Naviculaceae	Navicula	exigua var. signata	3
Pennales	Naviculaceae	Navicula	exilis	
Naviculales	Naviculaceae	Navicula	expecta	2
Pennales	Naviculaceae	Navicula	germainii	
Naviculales	Naviculaceae	Navicula	gibbosa	0
Naviculales	Naviculaceae	Navicula	gottlandica	2
Naviculales	Naviculaceae	Navicula	gregaria	2
Naviculales	Naviculaceae	Navicula	gysingensis	0
Pennales	Naviculaceae	Navicula	harderii	
Naviculales	Naviculaceae	Navicula	hasta	2
Naviculales	Naviculaceae	Navicula	hustedtii	3
Pennales	Naviculaceae	Navicula	incertata	
Naviculales	Naviculaceae	Navicula	ingenua	0
Naviculales	Naviculaceae	Navicula	integra	0
Pennales	Naviculaceae	Navicula	kotschyi	
Naviculales	Naviculaceae	Navicula	lanceolata	2
Naviculales	Naviculaceae	Navicula	lanceolata var. avenacea	2
Naviculales	Naviculaceae	Navicula	lateropunctata	0
Naviculales	Naviculaceae	Navicula	laterorostrata	1
Pennales	Naviculaceae	Navicula	lenzii	
Pennales	Naviculaceae	Navicula	libonensis	
Pennales	Naviculaceae	Navicula	longicephala	

Order	Family	Genus	Species	KPTI Value
Naviculales	Naviculaceae	Navicula	menisculus	2
Pennales	Naviculaceae	Navicula	menisculus var. obtusa	
Naviculales	Naviculaceae	Navicula	menisculus var. upsaliensis	2
Pennales	Naviculaceae	Navicula	microcephala	
Naviculales	Naviculaceae	Navicula	minima	1
Naviculales	Naviculaceae	Navicula	miniscula	0
Naviculales	Naviculaceae	Navicula	notha	3
Pennales	Naviculaceae	Navicula	occulta	
Naviculales	Naviculaceae	Navicula	paucivisitata	0
Pennales	Naviculaceae	Navicula	perminuta	
Pennales	Naviculaceae	Navicula	phyllepta	
Naviculales	Naviculaceae	Navicula	placenta	0
Naviculales	Naviculaceae	Navicula	placentula	0
Naviculales	Naviculaceae	Navicula	pseudanglica var. signata	0
Naviculales	Naviculaceae	Navicula	pseudarvensis	0
Naviculales	Naviculaceae	Navicula	pseudolanceolata	0
Naviculales	Naviculaceae	Navicula	radiosa	3
Naviculales	Naviculaceae	Navicula	radiosa var. parva	3
Naviculales	Naviculaceae	Navicula	recens	2
Pennales	Naviculaceae	Navicula	reichardtiana	
Naviculales	Naviculaceae	Navicula	rhynchocephala	3
Pennales	Naviculaceae	Navicula	rostellata	
Naviculales	Naviculaceae	Navicula	salinarum	2
Pennales	Naviculaceae	Navicula	salinicola	
Naviculales	Naviculaceae	Navicula	savannahiana	0
Pennales	Naviculaceae	Navicula	schadei	
Naviculales	Naviculaceae	Navicula	schroeteri	3
Naviculales	Naviculaceae	Navicula	scutelloides	0
Naviculales	Naviculaceae	Navicula	sp.	2
Pennales	Naviculaceae	Navicula	subminuscula	1
Pennales	Naviculaceae	Navicula	symmetrica	2
Pennales	Naviculaceae	Navicula	tantula	1
Naviculales	Naviculaceae	Navicula	tenelloides	3
Pennales	Naviculaceae	Navicula	terminata	
Pennales	Naviculaceae	Navicula	thienemannii	
Naviculales	Naviculaceae	Navicula	tridentula	0
Naviculales	Naviculaceae	Navicula	tripunctata	3
Naviculales	Naviculaceae	Navicula	trivialis	2
Naviculales	Naviculaceae	Navicula	vanheurckii	0

Order	Family	Genus	Species	KPTI Value
Naviculales	Naviculaceae	Navicula	veneta	1
Pennales	Naviculaceae	Navicula	viridula	2
Naviculales	Naviculaceae	Navicula	viridula var. linearis	2
Naviculales	Naviculaceae	Navicula	viridula var. rostellata	2
Pennales	Naviculaceae	Navicula	viridulacalcis	
Pennales	Naviculaceae	Navicula	wallacei	
Naviculales	Naviculaceae	Navicula	zanoni	2
Pennales	Neidiaceae	Nedium	ampliatum	
Naviculales	Neidiaceae	Neidium	affine	3
Naviculales	Neidiaceae	Neidium	affine var. amphirhynchus	3
Naviculales	Neidiaceae	Neidium	affine var. longiceps	3
Naviculales	Neidiaceae	Neidium	affinevar. undulatum	3
Naviculales	Neidiaceae	Neidium	alpinum	3
Naviculales	Neidiaceae	Neidium	apiculatum	0
Naviculales	Neidiaceae	Neidium	binodis	0
Naviculales	Neidiaceae	Neidium	bisulcatum	0
Naviculales	Neidiaceae	Neidium	bisulcatum var. baicalense	0
Naviculales	Neidiaceae	Neidium	dubium	0
Naviculales	Neidiaceae	Neidium	dubium f. constrictum	0
Naviculales	Neidiaceae	Neidium	iridis	0
Naviculales	Neidiaceae	Neidium	ladogense var. densestriatum	0
Naviculales	Neidiaceae	Neidium	sp.	3
Bacillariales	Bacillariaceae	Nitzschia	accomoda	0
Bacillariales	Bacillariaceae	Nitzschia	acicula	0
Bacillariales	Bacillariaceae	Nitzschia	aciculariodies	2
Bacillariales	Bacillariaceae	Nitzschia	acicularis	2
Pennales	Bacillariaceae	Nitzschia	acidoclinata	
Bacillariales	Bacillariaceae	Nitzschia	adapta	2
Pennales	Bacillariaceae	Nitzschia	aequorea	0
Bacillariales	Bacillariaceae	Nitzschia	agnita	3
Bacillariales	Bacillariaceae	Nitzschia	alpina	1
Bacillariales	Bacillariaceae	Nitzschia	amphibia	1
Pennales	Bacillariaceae	Nitzschia	angustata	
Bacillariales	Bacillariaceae	Nitzschia	angustata var. acuta	2
Bacillariales	Bacillariaceae	Nitzschia	angustatula	2
Bacillariales	Bacillariaceae	Nitzschia	angustula	2
Pennales	Bacillariaceae	Nitzschia	bacillum	
Pennales	Bacillariaceae	Nitzschia	bremensis	
Bacillariales	Bacillariaceae	Nitzschia	brevissima	0

Order	Family	Genus	Species	KPTI Value
Bacillariales	Bacillariaceae	Nitzschia	capitellata	1
Bacillariales	Bacillariaceae	Nitzschia	clausii	2
Pennales	Bacillariaceae	Nitzschia	closterium	
Bacillariales	Bacillariaceae	Nitzschia	communis	1
Bacillariales	Bacillariaceae	Nitzschia	compressa	0
Bacillariales	Bacillariaceae	Nitzschia	debilis	0
Bacillariales	Bacillariaceae	Nitzschia	denticula	3
Pennales	Bacillariaceae	Nitzschia	desertorum	
Bacillariales	Bacillariaceae	Nitzschia	dissipata	3
Bacillariales	Bacillariaceae	Nitzschia	dissipata var. media	3
Bacillariales	Bacillariaceae	Nitzschia	dubia	2
Bacillariales	Bacillariaceae	Nitzschia	elegantula	0
Bacillariales	Bacillariaceae	Nitzschia	filiformis	1
Bacillariales	Bacillariaceae	Nitzschia	filiformis var. conferta	
Bacillariales	Bacillariaceae	Nitzschia	fonticola	2
Bacillariales	Bacillariaceae	Nitzschia	frustulum	1
Bacillariales	Bacillariaceae	Nitzschia	frustulum var. perpusilla	1
Bacillariales	Bacillariaceae	Nitzschia	gandersheimiensis	0
Bacillariales	Bacillariaceae	Nitzschia	gracilis	2
Bacillariales	Bacillariaceae	Nitzschia	hantzschiana	0
Bacillariales	Bacillariaceae	Nitzschia	heufleriana	3
Bacillariales	Bacillariaceae	Nitzschia	inconspicua	2
Bacillariales	Bacillariaceae	Nitzschia	intermedia	2
Pennales	Bacillariaceae	Nitzschia	lanceolata	
Pennales	Bacillariaceae	Nitzschia	liebethruthii	
Bacillariales	Bacillariaceae	Nitzschia	linearis	3
Pennales	Bacillariaceae	Nitzschia	linearis var. subtilis	
Bacillariales	Bacillariaceae	Nitzschia	linearis var. tenuis	3
Bacillariales	Bacillariaceae	Nitzschia	lorenziana	3
Bacillariales	Bacillariaceae	Nitzschia	microcephala	1
Bacillariales	Bacillariaceae	Nitzschia	nana	3
Bacillariales	Bacillariaceae	Nitzschia	obtusatum	0
Bacillariales	Bacillariaceae	Nitzschia	palea	1
Bacillariales	Bacillariaceae	Nitzschia	palea var. debilis	1
Bacillariales	Bacillariaceae	Nitzschia	palea var. tenuirostris	1
Bacillariales	Bacillariaceae	Nitzschia	paleacea	2
Pennales	Bacillariaceae	Nitzschia	paleaeformis	
Bacillariales	Bacillariaceae	Nitzschia	parvula	0
Bacillariales	Bacillariaceae	Nitzschia	pellucida	0

Order	Family	Genus	Species	KPTI Value
Bacillariales	Bacillariaceae	Nitzschia	perminuta	2
Bacillariales	Bacillariaceae	Nitzschia	pumila	2
Pennales	Bacillariaceae	Nitzschia	pura	
Bacillariales	Bacillariaceae	Nitzschia	pusilla	1
Bacillariales	Bacillariaceae	Nitzschia	radicula	0
Bacillariales	Bacillariaceae	Nitzschia	rautenbachiae	3
Bacillariales	Bacillariaceae	Nitzschia	recta	3
Bacillariales	Bacillariaceae	Nitzschia	reversa	2
Bacillariales	Bacillariaceae	Nitzschia	romana	3
Bacillariales	Bacillariaceae	Nitzschia	rostellata	0
Bacillariales	Bacillariaceae	Nitzschia	sigma	1
Bacillariales	Bacillariaceae	Nitzschia	sigmoidea	3
Bacillariales	Bacillariaceae	Nitzschia	sinuata var. delognei	2
Pennales	Bacillariaceae	Nitzschia	sinuata var. tabellaria	3
Bacillariales	Bacillariaceae	Nitzschia	sociabilis	2
Pennales	Bacillariaceae	Nitzschia	solita	
Bacillariales	Bacillariaceae	Nitzschia	sp.	2
Bacillariales	Bacillariaceae	Nitzschia	stricta	0
Bacillariales	Bacillariaceae	Nitzschia	subacicularis	0
Pennales	Bacillariaceae	Nitzschia	subcapitellata	
Bacillariales	Bacillariaceae	Nitzschia	sublinearis	2
Pennales	Bacillariaceae	Nitzschia	suchlandtii	
Pennales	Bacillariaceae	Nitzschia	supralitorea	
Bacillariales	Bacillariaceae	Nitzschia	tropica	2
Pennales	Bacillariaceae	Nitzschia	tubicola	
Bacillariales	Bacillariaceae	Nitzschia	umbonata	0
Bacillariales	Bacillariaceae	Nitzschia	vermicularis	2
Bacillariales	Bacillariaceae	Nitzschia	vitrea	0
Pennales	Bacillariaceae	Nitzschia	wuellerstorffii	
Pennales	Naviculaceae	Nupela	impexiformis	
Pennales	Naviculaceae	Nupela	lapidosa	
Pennales	Naviculaceae	Nupela	neglecta	
Pennales	Naviculaceae	Nupela	silvahercynia	
Pennales	Naviculaceae	Nupela	sp.	
Pennales	Naviculaceae	Nupela	vitiosa	
Centrales	Orthoseiraceae	Orthoseira	roseana	0
Centrales	Orthoseiraceae	Orthoseira	sp.	
Pennales	Berkekeyaceae	Parlibellus	crucicula	
Naviculales	Pinnulariaceae	Pinnularia	acrosphaeria	0

Order	Family	Genus	Species	KPTI Value
Naviculales	Pinnulariaceae	Pinnularia	amphisbaena	0
Naviculales	Pinnulariaceae	Pinnularia	appendiculata	2
Naviculales	Pinnulariaceae	Pinnularia	bacillum	3
Pennales	Pinnulariaceae	Pinnularia	biceps	3
Naviculales	Pinnulariaceae	Pinnularia	borealis	2
Naviculales	Pinnulariaceae	Pinnularia	borealis var. rectangularis	2
Naviculales	Pinnulariaceae	Pinnularia	branderi	0
Naviculales	Pinnulariaceae	Pinnularia	braunii var. amphicephala	3
Naviculales	Pinnulariaceae	Pinnularia	budensis	0
Naviculales	Pinnulariaceae	Pinnularia	gibba	3
Naviculales	Pinnulariaceae	Pinnularia	gibba f. sumbundulata	3
Naviculales	Pinnulariaceae	Pinnularia	gibba var. mesogonglya	3
Naviculales	Pinnulariaceae	Pinnularia	gibba var. rostrata	3
Naviculales	Pinnulariaceae	Pinnularia	hyalina	0
Naviculales	Pinnulariaceae	Pinnularia	interrupta	3
Naviculales	Pinnulariaceae	Pinnularia	legumen	3
Pennales	Pinnulariaceae	Pinnularia	macilenta	
Naviculales	Pinnulariaceae	Pinnularia	maior	0
Pennales	Pinnulariaceae	Pinnularia	mesogongyla	3
Naviculales	Pinnulariaceae	Pinnularia	microstauron	0
Naviculales	Pinnulariaceae	Pinnularia	nodosa	0
Naviculales	Pinnulariaceae	Pinnularia	obscura	3
Naviculales	Pinnulariaceae	Pinnularia	schumanniana	0
Naviculales	Pinnulariaceae	Pinnularia	schumanniana var. bioconstricta	2
Naviculales	Pinnulariaceae	Pinnularia	schumanniana var. bioconstricta	2
Naviculales	Pinnulariaceae	Pinnularia	silicula	3
Naviculales	Pinnulariaceae	Pinnularia	silicula var truncatula	3
Naviculales	Pinnulariaceae	Pinnularia	silicula var. alpina	3
Naviculales	Pinnulariaceae	Pinnularia	silicula var. minuta	3
Naviculales	Pinnulariaceae	Pinnularia	silicula var. subundulata	3
Naviculales	Pinnulariaceae	Pinnularia	sp.	2
Naviculales	Pinnulariaceae	Pinnularia	sp.	3
Naviculales	Pinnulariaceae	Pinnularia	stomatophora	0
Naviculales	Pinnulariaceae	Pinnularia	streptoraphe	0
Naviculales	Pinnulariaceae	Pinnularia	subcapitata	3
Naviculales	Pinnulariaceae	Pinnularia	subcapitata var. paucistriata	3
Naviculales	Pinnulariaceae	Pinnularia	termitina	0
Naviculales	Pinnulariaceae	Pinnularia	thermalis	0
Naviculales	Pinnulariaceae	Pinnularia	undulata	2

Order	Family	Genus	Species	KPTI Value
Naviculales	Pinnulariaceae	Pinnularia	viridis	0
Pennales	Cymbellaceae	Placoneis	clementis	
Pennales	Cymbellaceae	Placoneis	clementoides	
Pennales	Cymbellaceae	Placoneis	elginensis	
Pennales	Cymbellaceae	Placoneis	exigua	
Cymbellales	Cymbellaceae	Placoneis	gastrum	0
Pennales	Cymbellaceae	Placoneis	placentula	
Pennales	Cymbellaceae	Placoneis	pseudanglica	
Naviculales	Plagiotropidaceae	Plagiotropis	lepidoptera var. proboscidea	3
Pennales	Achnanthaceae	Planothidium	daui	
Pennales	Achnanthaceae	Planothidium	delicatulum	
Pennales	Achnanthaceae	Planothidium	dispar	
Pennales	Achnanthaceae	Planothidium	distinctum	
Pennales	Achnanthaceae	Planothidium	dubium	
Pennales	Achnanthaceae	Planothidium	ellipticum	
Pennales	Achnanthaceae	Planothidium	engelbrechtii	
Pennales	Achnanthaceae	Planothidium	frequentissimum	
Pennales	Achnanthaceae	Planothidium	granum	
Pennales	Achnanthaceae	Planothidium	hauckianum	
Pennales	Achnanthaceae	Planothidium	haynaldii	
Pennales	Achnanthaceae	Planothidium	joursacense	
Acnanthales	Achnanthidiaceae	Planothidium	lanceolata	3
Acnanthales	Achnanthidiaceae	Planothidium	lanceolata var. apiculata	3
Acnanthales	Achnanthidiaceae	Planothidium	lanceolata var. dubia	3
Pennales	Achnanthaceae	Planothidium	lanceolatum	
Pennales	Achnanthaceae	Planothidium	rostratum	
Pennales	Achnanthaceae	Planothidium	stewartii	
Pennales	Achnanthaceae	Platessa	conspicua	
Pennales	Achnanthaceae	Platessa	holsatica	
Pennales	Achnanthaceae	Platessa	hustedtii	
Pennales	Achnanthaceae	Platessa	montana	
Pennales	Achnanthaceae	Platessa	rupestris	
Naviculales	Pleurosigmataceae	Pleurosigma	delicatulum	0
Triceratiales	Triceratiaceae	Pleurosira	laevis	3
Bacillariales	Bacillariaceae	Psammodictyon	constrictum	3
Pennales	Achnanthaceae	Psammothidium	bioretii	
Pennales	Achnanthaceae	Psammothidium	chlidanos	
Pennales	Achnanthaceae	Psammothidium	helveticum	
Pennales	Achnanthaceae	Psammothidium	marginulatum	

Order	Family	Genus	Species	KPTI Value
Pennales	Achnanthaceae	Psammothidium	subatomoides	
Pennales	Achnanthaceae	Psammothidium	ventralis	
Pennales	Fragilariaceae	Pseudostaurosira	binodis	0
Pennales	Fragilariaceae	Pseudostaurosira	brevistriata	0
Pennales	Fragilariaceae	Pseudostaurosira	parasitica	
Pennales	Fragilariaceae	Pseudostaurosira	parasitica var. subconstricta	
Pennales	Fragilariaceae	Pseudostaurosira	pseudoconstruens	
Fragilariales	Fragilariaceae	Punctastriata	pinnata	3
Cymbellales	Gomophonemataceae	Reimeria	sinuata	4
Cymbellales	Rhoicospheniaceae	Rhoicosphenia	abbreviata	3
Rhopalodiales	Rhopalodiaceae	Rhopalodia	gibba	3
Rhopalodiales	Rhopalodiaceae	Rhopalodia	gibba var. ventricosa	4
Rhopalodiales	Rhopalodiaceae	Rhopalodia	gibberula var. vanheurckii	0
Pennales	Achnanthaceae	Rossithidium	duthii	
Acnanthales	Achnanthidiaceae	Rossithidium	linearis	3
Acnanthales	Achnanthidiaceae	Rossithidium	linearis f. curta	3
Acnanthales	Achnanthidiaceae	Rossithidium	linearis var. pusilla	3
Pennales	Achnanthaceae	Rossithidium	petersennii	
Naviculales	Sellaphoraceae	Sellaphora	bacillum	4
Pennales	Sellaphoraceae	Sellaphora	hustedtii	
Naviculales	Sellaphoraceae	Sellaphora	laevissima	0
Pennales	Sellaphoraceae	Sellaphora	mutata	
Naviculales	Sellaphoraceae	Sellaphora	pupula	3
Naviculales	Sellaphoraceae	Sellaphora	pupula var. capitata	3
Naviculales	Sellaphoraceae	Sellaphora	pupula var. elliptica	3
Naviculales	Sellaphoraceae	Sellaphora	pupula var. mutata	3
Naviculales	Sellaphoraceae	Sellaphora	pupula var. rectangularis	3
Naviculales	Sellaphoraceae	Sellaphora	pupula var. rostrata	3
Naviculales	Sellaphoraceae	Sellaphora	pupula var. subcapitata	3
Pennales	Sellaphoraceae	Sellaphora	rostrata	
Naviculales	Sellaphoraceae	Sellaphora	seminulum	1
Naviculales	Sellaphoraceae	Sellaphora	seminulum var hustedtii	1
Bacillariales	Bacillariaceae	Simonsenia	delognei	3
Centrales	Skeletonemaceae	Skeletonema	potomos	3
Pennales	Stauroneidaceae	Stauroneis	agrestis	
Naviculales	Stauroneidaceae	Stauroneis	anceps	4
Naviculales	Stauroneidaceae	Stauroneis	anceps var. american	4
Naviculales	Stauroneidaceae	Stauroneis	anceps var. gracilis	4
Naviculales	Stauroneidaceae	Stauroneis	anceps var. linearis	4

Order	Family	Genus	Species	KPTI Value
Naviculales	Stauroneidaceae	Stauroneis	kriegeri	0
Naviculales	Stauroneidaceae	Stauroneis	legumen	0
Naviculales	Stauroneidaceae	Stauroneis	nana	0
Naviculales	Stauroneidaceae	Stauroneis	nobilis	0
Naviculales	Stauroneidaceae	Stauroneis	obtusa	0
Naviculales	Stauroneidaceae	Stauroneis	phoenicenteron	0
Naviculales	Stauroneidaceae	Stauroneis	phoenicenteron f. gracilis	0
Naviculales	Stauroneidaceae	Stauroneis	smithii	4
Naviculales	Stauroneidaceae	Stauroneis	smithii var. incisa	4
Naviculales	Stauroneidaceae	Stauroneis	smithii var. sagitta	4
Naviculales	Stauroneidaceae	Stauroneis	sp.	4
Pennales	Stauroneidaceae	Stauroneis	thermicola	
Pennales	Fragilariaceae	Staurosira	construens	0
Pennales	Fragilariaceae	Staurosira	construens f. Venter	0
Pennales	Fragilariaceae	Staurosira	construens var. binodis	
Pennales	Fragilariaceae	Staurosira	construens var. subsalina	0
Pennales	Fragilariaceae	Staurosira	construens var. venter	0
Pennales	Fragilariaceae	Staurosira	elliptica	
Pennales	Fragilariaceae	Staurosirella	lapponica	0
Pennales	Fragilariaceae	Staurosirella	leptostauron	3
Pennales	Fragilariaceae	Staurosirella	leptostauron var. dubia	
Pennales	Fragilariaceae	Staurosirella	leptostauron var. rhomboides	
Pennales	Fragilariaceae	Staurosirella	pinnata	
Pennales	Fragilariaceae	Staurosirella	pinnata var. intercedens	
Pennales	Surirellaceae	Stenopterobia	curvula	
Surirellales	Surirellaceae	Stenopterobia	delicatissima	4
Thalassiosirales	Stephanodiscaceae	Stephanocyclus	meneghiniana	1
Centrales	Stephanodiscaceae	Stephanodiscus	alpinus	0
Centrales	Stephanodiscaceae	Stephanodiscus	hantzschii	3
Centrales	Stephanodiscaceae	Stephanodiscus	minutulus	3
Centrales	Stephanodiscaceae	Stephanodiscus	niagarae	0
Centrales	Stephanodiscaceae	Stephanodiscus	sp.	3
Centrales	Stephanodiscaceae	Stephanodiscus	subtilis	0
Centrales	Stephanodiscaceae	Stephanodiscus	tenuis	3
Surirellales	Surirellaceae	Surirella	agmatilis	3
Pennales	Surirellaceae	Surirella	amphioxys	
Pennales	Surirellaceae	Surirella	angusta	
Pennales	Surirellaceae	Surirella	angusta	
Surirellales	Surirellaceae	Surirella	angustata var. acuta	2

Order	Family	Genus	Species	KPTI Value
Surirellales	Surirellaceae	Surirella	brebissonii	0
Surirellales	Surirellaceae	Surirella	elegans	4
Surirellales	Surirellaceae	Surirella	gracilis	0
Surirellales	Surirellaceae	Surirella	linearis	2
Surirellales	Surirellaceae	Surirella	linearis var. helvetica	2
Surirellales	Surirellaceae	Surirella	minuta	2
Surirellales	Surirellaceae	Surirella	minuta var. africana	2
Surirellales	Surirellaceae	Surirella	minuta var. pinnata	3
Surirellales	Surirellaceae	Surirella	ovalis	3
Pennales	Surirellaceae	Surirella	patella	
Pennales	Surirellaceae	Surirella	roba	
Surirellales	Surirellaceae	Surirella	robusta	0
Surirellales	Surirellaceae	Surirella	robusta f. lata	0
Surirellales	Surirellaceae	Surirella	sp.	2
Surirellales	Surirellaceae	Surirella	splendida	0
Surirellales	Surirellaceae	Surirella	tenera	3
Surirellales	Surirellaceae	Surirella	tenera var. nervosa	3
Pennales	Surirellaceae	Surirella	tenuis	
Pennales	Fragilariaceae	Synedra	acus	3
Pennales	Fragilariaceae	Synedra	delicatissima	
Pennales	Fragilariaceae	Synedra	filiformis var. exilis	4
Pennales	Fragilariaceae	Synedra	goulardi	
Pennales	Fragilariaceae	Synedra	mazamaensis	
Pennales	Fragilariaceae	Synedra	nanana	4
Pennales	Fragilariaceae	Synedra	rumpens	4
Pennales	Fragilariaceae	Synedra	rumpes var. fragilaroides	4
Pennales	Fragilariaceae	Synedra	sp.	3
Pennales	Fragilariaceae	Synedra	ulna	3
Pennales	Fragilariaceae	Synedra	ulna var. ramesi	3
Pennales	Tabellariaceae	Tabellaria	fenestrata	4
Pennales	Tabellariaceae	Tabellaria	flocculosa	4
Pennales	Tabellariaceae	Tabellaria	quadriseptata	
Pennales	Fragilariaceae	Tabularia	fasciculata	
Pennales	Tabellariaceae	Tetracyclus	glans	0
Pennales	Tabellariaceae	Tetracyclus	rupestris	0
Centrales	Thalassiosiraceae	Thalassiosira	bramaputrae	
Centrales	Thalassiosiraceae	Thalassiosira	pseudonana	
Centrales	Thalassiosiraceae	Thalassiosira	visurgis	
Centrales	Thallassiosiraceae	Thalassiosira	weissflogii	2

Order	Family	Genus	Species	KPTI Value
Pennales	Bacillariaceae	Tryblionella	aerophila	
Bacillariales	Bacillariaceae	Tryblionella	apiculata	1
Pennales	Bacillariaceae	Tryblionella	calida	
Bacillariales	Bacillariaceae	Tryblionella	coarctata	3
Pennales	Bacillariaceae	Tryblionella	compressa	
Pennales	Bacillariaceae	Tryblionella	debilis	
Bacillariales	Bacillariaceae	Tryblionella	gracilis	3
Bacillariales	Bacillariaceae	Tryblionella	hungarica	2
Bacillariales	Bacillariaceae	Tryblionella	levidensis	3
Bacillariales	Bacillariaceae	Tryblionella	littoralis	3
Bacillariales	Bacillariaceae	Tryblionella	victoriae	3

NON-DIATOM TAXA LIST (Sorted by Genus)

Phylum	Class	Order	Family	Genus	Species
Cyanophyta	Myxophyceae	Nostocales	Nostocaceae	Anabaena	subcylindrica
Chlorophyta	Chlorophyceae	Oedogoniales	Oedogoniaceae	Bulbocheate	sp.
Cyanophyta	Myxophyceae	Nostocales	Rivulariaceae	Calothrix	sp.
Cyanophyta	Myxophyceae	Chrococcales	Chamaesiphonaceae	Chamaesiphon	confervicolus
Cyanophyta	Myxophyceae	Chrococcales	Chroococcaceae	Chrococcus	distans
Cyanophyta	Myxophyceae	Chrococcales	Chroococcaceae	Chrococcus	limneticus
Cyanophyta	Myxophyceae	Chrococcales	Chroococcaceae	Chrococcus	sp.
Chlorophyta	Chlorophyceae	Cladophorales	Cladophoraceae	Cladophora	glomerata
Chlorophyta	Chlorophyceae	Chlorococcales	Oocystaceae	Closteriopsis	longissima
Chlorophyta	Chlorophyceae	Zygnematales	Desmidiaceae	Closterium	lunula
Chlorophyta	Chlorophyceae	Zygnematales	Desmidiaceae	Closterium	moniliferum
Chlorophyta	Chlorophyceae	Zygnematales	Desmidiaceae	Closterium	sp.
Chlorophyta	Chlorophyceae	Zygnematales	Desmidiaceae	Cosmarium	dentatum
Chlorophyta	Chlorophyceae	Zygnematales	Desmidiaceae	Cosmarium	garrolense
Chlorophyta	Chlorophyceae	Zygnematales	Desmidiaceae	Cosmarium	garrolense var. crassum
Chlorophyta	Chlorophyceae	Zygnematales	Desmidiaceae	Cosmarium	sp.
Cryptophyta	Cryptophyceae	Cryptomonadales	Cryptomonadaceae	Cryptomonas	sp.
Cryptophyta	Cryptophyceae	Not Designated	Not Designated	Cryptophyte	alga
Chlorophyta	Chlorophyceae	Zygnematales	Desmidiaceae	Desmidium	grevellii
Chlorophyta	Chlorophyceae	Volvocales	Volvocaceae	Eudorina	elegans
Euglenophyta	Euglenophyceae	Euglenales	Euglenaceae	Euglena	acus
Euglenophyta	Euglenophyceae	Euglenales	Euglenaceae	Euglena	minuta
Euglenophyta	Euglenophyceae	Euglenales	Euglenaceae	Euglena	sp.
Euglenophyta	Euglenophyceae	Euglenales	Euglenaceae	Euglena	spirogyra
Cyanophyta	Myxophyceae	Oscillatoriales	Pseudanabaenaceae	Geitlerinema	splendidum
Cyanophyta	Myxophyceae	Oscillatoriales	Homoeotrichaceae	Heteroleibleinia	kuetzingii
Cyanophyta	Myxophyceae	Oscillatoriales	Homoeotrichaceae	Heteroleibleinia	sp.
Cyanophyta	Myxophyceae	Oscillatoriales	Pseudanabaenaceae	Homoeothrix	hansgirgi
Cyanophyta	Myxophyceae	Oscillatoriales	Pseudanabaenaceae	Homoeothrix	janthina
Cyanophyta	Myxophyceae	Oscillatoriales	Pseudanabaenaceae	Homoeothrix	juliana
Cyanophyta	Myxophyceae	Oscillatoriales	Pseudanabaenaceae	Homoeothrix	simplex
Cyanophyta	Myxophyceae	Oscillatoriales	Pseudanabaenaceae	Homoeothrix	sp.
Cyanophyta	Myxophyceae	Oscillatoriales	Pseudanabaenaceae	Homoeothrix	varians
Chlorophyta	Chlorophyceae	Chlorococcales	Hydrodictyaceae	Hydrodictyon	reticulatum
Chlorophyta	Chlorophyceae	Chlorococcales	Oocystaceae	Kirchneriella	contorta
Cyanophyta	Myxophyceae	Oscillatoriales	Borziaceae	Komvophoron	schmidlei
Cyanophyta	Myxophyceae	Oscillatoriales	Borziaceae	Komvophoron	sp.

Phylum	Class	Order	Family	Genus	Species
Cyanophyta	Myxophyceae	Oscillatoriales	Pseudanabaenaceae	Leptolyngbya	angustissima
Cyanophyta	Myxophyceae	Oscillatoriales	Pseudanabaenaceae	Leptolyngbya	sp.
Cyanophyta	Myxophyceae	Oscillatoriales	Pseudanabaenaceae	Lyngbya	epiphytica
Cyanophyta	Myxophyceae	Oscillatoriales	Oscillatoriaceae	Lyngbya	martensiana
Cyanophyta	Myxophyceae	Chroococcales	Merismopediaceae	Merismopedia	glauca
Cyanophyta	Myxophyceae	Chroococcales	Merismopediaceae	Merismopedia	tenuissima
Chlorophyta	Clorophyceae	Zygnematales	Desmidaceae	Microasterias	radiosa
Cyanophyta	Myxophyceae	Oscillatoriales	Phormidaceae	Microcoleus	vaginatus
Chlorophyta	Chlorophyceae	Chlorococcales	Microsporaceae	Microspora	amoena
Chlorophyta	Chlorophyceae	Zygnematales	Zygnemataceae	Mougeotia	sp.
Chlorophyta	Chlorophyceae	Oedogoniales	Oedogoniaceae	Oedogonium	sp.
Chlorophyta	Chlorophyceae	Chlorococales	Oocystaceae	Oocystis	parva
Cyanophyta	Myxophyceae	Oscillatoriales	Oscillatoriaceae	Oscillatoria	limosa
Cyanophyta	Myxophyceae	Oscillatoriales	Oscillatoriaceae	Oscillatoria	retzii
Cyanophyta	Myxophyceae	Oscillatoriales	Oscillatoriaceae	Oscillatoria	sp.
Chlorophyta	Chlorophyceae	Chlorococcales	Hydrodictyaceae	Pediastrum	biradiatum
Chlorophyta	Chlorophyceae	Chlorococcales	Hydrodictyaceae	Pediastrum	boryanum
Pyrrhophyta	Dinophyceae	Peridiniales	Peridiniaceae	Peridiniopsis	sp.
Euglenophyta	Euglenophyceae	Euglenales	Euglenaceae	Phacus	longicauda
Euglenophyta	Euglenophyceae	Euglenales	Euglenaceae	Phacus	orbicularis
Cyanophyta	Myxophyceae	Oscillatoriales	Phormidiaceae	Phormidium	acutissimum
Cyanophyta	Myxophyceae	Oscillatoriales	Oscillatoriales	Phormidium	aerugineo-caeruleum
Cyanophyta	Myxophyceae	Oscillatoriales	Phormidiaceae	Phormidium	ambiguum
Cyanophyta	Myxophyceae	Oscillatoriales	Phormidiaceae	Phormidium	amoenum
Cyanophyta	Myxophyceae	Oscillatoriales	Phormidiaceae	Phormidium	autumnale
Cyanophyta	Myxophyceae	Oscillatoriales	Oscillatoriales	Phormidium	breve
Cyanophyta	Myxophyceae	Oscillatoriales	Phormidiaceae	Phormidium	caeruleum
Cyanophyta	Myxophyceae	Oscillatoriales	Phormidiaceae	Phormidium	chalybeum
Cyanophyta	Myxophyceae	Oscillatoriales	Phormidiaceae	Phormidium	granulatum
Cyanophyta	Myxophyceae	Oscillatoriales	Phormidiaceae	Phormidium	minnesotense
Cyanophyta	Myxophyceae	Oscillatoriales	Phormidiaceae	Phormidium	retzii
Cyanophyta	Myxophyceae	Oscillatoriales	Phormidiaceae	Phormidium	sp.
Cyanophyta	Myxophyceae	Oscillatoriales	Phormidiaceae	Phormidium	tenue
Cyanophyta	Myxophyceae	Oscillatoriales	Phormidiaceae	Planktothrix	prolifica
Cyanophyta	Myxophyceae	Oscillatoriales	Pseudanabaenaceae	Pseudanabaena	sp.
Chlorophyta	Chlorophyceae	Cladophorales	Cladophoraceae	Rhizoclonium	sp.
Chlorophyta	Chlorophyceae	Chlorococcales	Scenedesmaceae	Scenedesmus	acuminatus
Chlorophyta	Chlorophyceae	Chlorococcales	Scenedesmaceae	Scenedesmus	bijuga

Phylum	Class	Order	Family	Genus	Species
Chlorophyta	Chlorophyceae	Chlorococcales	Scenedesmaceae	Scenedesmus	dimorphus
Chlorophyta	Chlorophyceae	Chlorococcales	Scenedesmaceae	Scenedesmus	ecornis
Chlorophyta	Chlorophyceae	Chlorococcales	Scenedesmaceae	Scenedesmus	sp.
Chlorophyta	Chlorophyceae	Chlorococcales	Scenedesmaceae	Scenedesmus	spinosus
Chlorophyta	Chlorophyceae	Zygnematales	Zygnemataceae	Spirogyra	sp.
Chlorophyta	Chlorophyceae	Zygnematales	Desmidiaceae	Staurastrum	punctulatum
Chlorophyta	Chlorophyceae	Zygnematales	Desmidiaceae	Staurastrum	sp.
Chlorophyta	Chlorophyceae	Chaetophorales	Chaetophoracea	Stigeoclonium	lubricum
Euglenophyta	Euglenophyceae	Euglenales	Euglenaceae	Strombomonas	deflandrei
Euglenophyta	Euglenophyceae	Euglenales	Euglenaceae	Strombomonas	sp.
Chlorophytae	Chlorophyceae	Chlorococcales	Chlorococcaceae	Tetraedron	minimum
Chlorophytae	Chlorophyceae	Chlorococcales	Chlorococcaceae	Tetraedron	muticum
Euglenophyta	Euglenophyceae	Euglenales	Euglenaceae	Trachelomonas	hispida
Euglenophyta	Euglenophyceae	Euglenales	Euglenaceae	Trachelomonas	ovata
Euglenophyta	Euglenophyceae	Euglenales	Euglenaceae	Trachelomonas	sp.
Chrysophyta	Xanthophyceae	Heterotrichales	Tribonemataceae	Tribonema	sp.
Chlorophytae	Chlorophyceae	Ulotrichales	Ulotrichaceae	Ulothrix	tenuissima
Chlorophyta	Chlorophyceae	Ulotrichales	Ulotrichaceae	Ulothrix	variabilis
Chlorophyta	Chlorophyceae	Ulotrichales	Ulotrichaceae	Ulothrix	zonata
Chrysophyta	Xanthophyceae	Vaucheriales	Vaucheriaceae	Vaucheria	sp.

APPENDIX E

BIOVOLUME CALCULATION

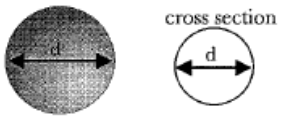
GEOMETRIC SHAPES AND EQUATIONS FOR CALCULATION OF BIOVOLUME
MODELS FOR BIOVOLUME CALCULATIONS OF MICROALGAL TAXA

GEOMETRIC SHAPES AND EQUATIONS FOR CALCULATION OF BIOVOLUME

From Hillebrand et al, 1999

TABLE 1. Geometric shapes and equations for the calculation of biovolume. Shapes are drawn in a three-dimensional version and in cross sections. Equations are given, using standard abbreviations for the linear dimensions to be measured. Abbreviations: A = surface area; V = volume; r = radius; d = diameter; h = height; a = apical axis (length); b = transapical axis (width); c = pervalvar axis (height); z = height of cone; l = length of one side; m = height of a triangle.

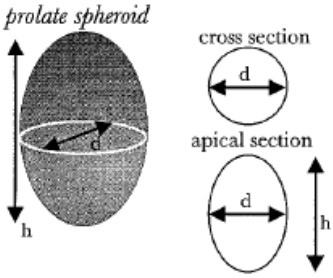
sphere



$$V = \frac{4}{3} \cdot \pi \cdot r^3 = \frac{\pi}{6} \cdot d^3$$

$$A = 4 \cdot \pi \cdot r^2 = \pi \cdot d^2$$

prolate spheroid

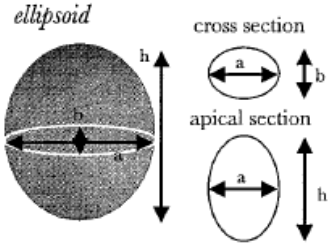


Subspherical body with circular cross-section and elliptical apical section. Surface area is given for $\sin^{-1}$ measured in radians and for h>d.

$$V = \frac{\pi}{6} \cdot d^2 \cdot h$$

$$A = \frac{\pi \cdot d}{2} \cdot \left(d + \frac{h^2}{\sqrt{h^2 - d^2}} \sin^{-1} \frac{\sqrt{h^2 - d^2}}{h} \right)$$

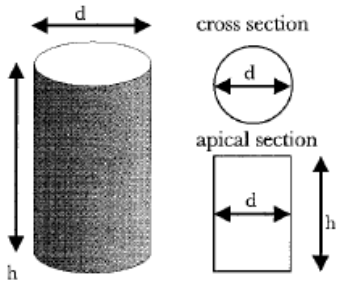
ellipsoid



This body is subspherical with three different dimensions (prolate spheroid with elliptical cross-sections).

$$V = \frac{\pi}{6} \cdot a \cdot b \cdot h$$

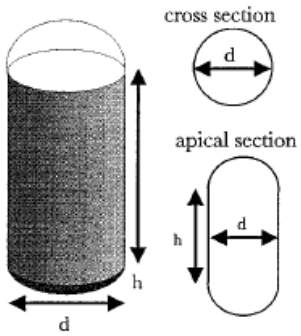
cylinder



$$V = \pi \cdot r^2 \cdot h = \frac{\pi}{4} \cdot d^2 \cdot h$$

$$A = 2 \cdot \pi \cdot r^2 + 2 \cdot \pi \cdot r \cdot h = \pi \cdot d \cdot \left(\frac{d}{2} + h\right)$$

cylinder + 2 half spheres



This body refers mostly to cylindric diatoms with domed valves such as *Stephanopyxis*.

$$V = \pi \cdot r^2 \cdot h + \frac{4}{3} \cdot \pi \cdot r^2$$

$$= \frac{1}{4}\pi \cdot d^2 \cdot h + \frac{1}{6} \cdot \pi \cdot d^3$$

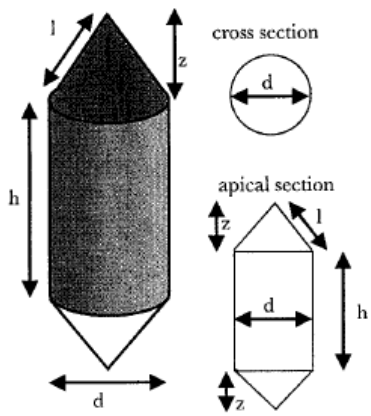
$$= \pi \cdot d^2 \cdot \left(\frac{h}{4} + \frac{d}{6}\right)$$

$$A = 4 \cdot \pi \cdot r^2 + 2 \cdot \pi \cdot r \cdot h = \pi \cdot d \cdot (d + h)$$

GEOMETRIC SHAPES AND EQUATIONS FOR CALCULATION OF BIOVOLUME (cont.)

TABLE 1. Continued.

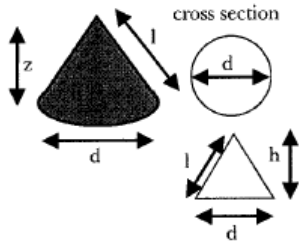
cylinder + 2 cones



Similar to the previous shape this body refers to cylindric species, but here the cells have acute apices.

$$V = \frac{\pi}{4} \cdot d^2 \cdot h + 2 \cdot \frac{\pi}{12} \cdot d^2 \cdot z$$
$$= \frac{\pi}{4} \cdot d^2 \cdot \left(h + \frac{z}{2}\right)$$
$$A = d \cdot \pi \cdot h + \pi \cdot d \cdot l = \pi \cdot d \cdot (h + l)$$

cone

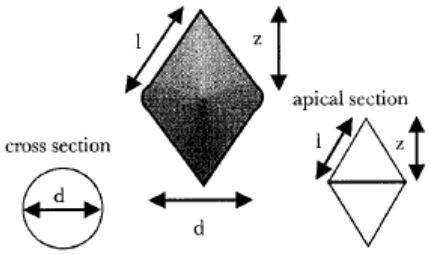


$$V = \tfrac{1}{3} \cdot \pi \cdot r^2 \cdot z = \tfrac{\pi}{12} \cdot d^2 \cdot z$$
$$A = \pi \cdot r^2 + \pi \cdot r \cdot l = \tfrac{\pi}{2} \cdot d \cdot (\tfrac{d}{2} + l)$$

For truncated cones (e.g. several species of the Desmidiaceae) the following equations can be used (note: the subscripts 1 and 2 refer to the upper and lower radius and diameter of the truncated cone):

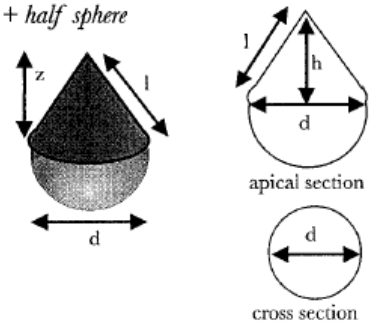
$$V = \frac{1}{3} \cdot \pi \cdot z \cdot (r_1^2 + r_1 \cdot r_2 + r_2^2)$$
$$= \frac{\pi}{12} \cdot z \cdot (d_1^2 + d_1 \cdot d_2 + d_2^2)$$
$$A = \tfrac{\pi}{4} \cdot (d_2^2 + d_1^2 + 2l[d_2 + d_1])$$

double cone



$$V = 2 \cdot \tfrac{1}{3} \cdot \pi \cdot r^2 \cdot z = \tfrac{\pi}{6} \cdot d^2 \cdot z$$
$$A = 2 \cdot \pi \cdot r \cdot l = \pi \cdot d \cdot l$$

cone + half sphere

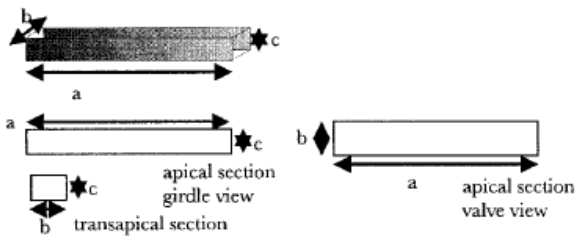


$$V = \frac{1}{3} \cdot \pi \cdot r^2 \cdot z + \frac{1}{2} \cdot \frac{4}{3} \cdot \pi \cdot r^3$$
$$= \frac{\pi}{12} \cdot d^2 \cdot (z + d)$$
$$A = \pi \cdot r \cdot l + 2 \cdot \pi \cdot r^2$$
$$= \frac{1}{2}\pi \cdot d \cdot l + \frac{1}{2}\pi \cdot d^2$$
$$= \frac{1}{2}\pi \cdot d \cdot (l + d)$$

GEOMETRIC SHAPES AND EQUATIONS FOR CALCULATION OF BIOVOLUME (cont.)

TABLE 1. Continued.

rectangular box

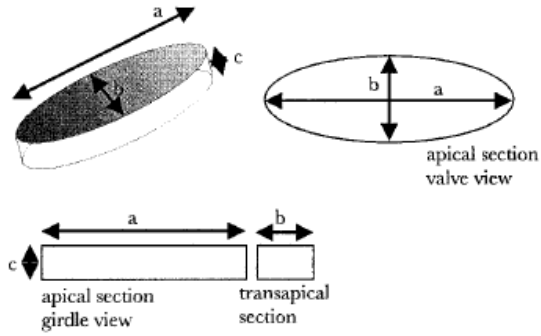


$$V = a \cdot b \cdot c$$

$$A = 2 \cdot a \cdot b + 2 \cdot b \cdot c + 2 \cdot a \cdot c$$

A cube is a special case of this shape where a=b=c, then $V=a^3$ and $A=6 \cdot a^2$.

prism on elliptic base

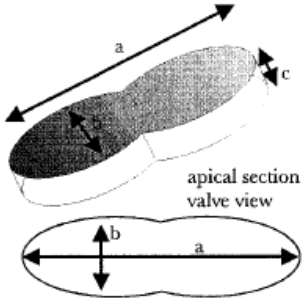


This shape is suitable for elliptic pennate diatoms, even if they are constricted in valve view - then the mean of both the central width and maximum width is taken.

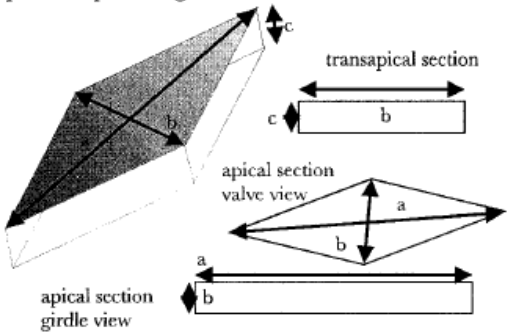
$$V = \frac{\pi}{4} \cdot a \cdot b \cdot c$$

$$A = 2 \cdot \frac{1}{4} \cdot \pi \cdot a \cdot b + \frac{1}{2} \cdot \pi \cdot (a+b) \cdot c = \frac{\pi}{2} \cdot (a \cdot b + [a+b] \cdot c)$$

elliptic prism with transapical constriction



prism on parallelogram-base



Rhombic diatom species belong for example to the genera *Pleurosigma* and *Gyrosigma*, the basic parallelogram is even-sided.

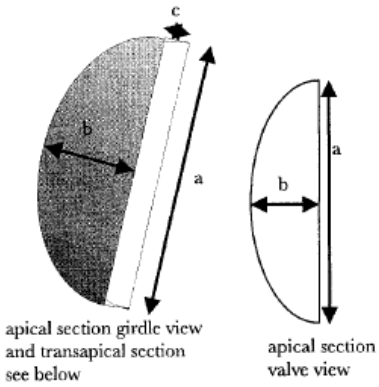
$$V = \frac{1}{2} \cdot a \cdot b \cdot c$$

$$A = a \cdot b + \frac{\sqrt{a^2+b^2}}{4} \cdot c$$

GEOMETRIC SHAPES AND EQUATIONS FOR CALCULATION OF BIOVOLUME (cont.)

TABLE 1. Continued.

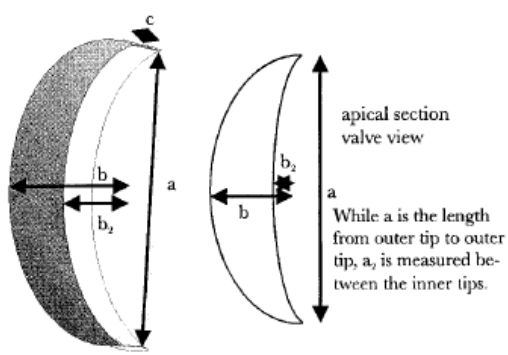
half-elliptic prism



$$V = \frac{1}{2} \cdot \frac{1}{4}\pi \cdot a \cdot 2b \cdot c = \frac{\pi}{4} \cdot a \cdot b \cdot c$$

$$A = 2 \cdot \frac{1}{4} \cdot \frac{1}{2} \cdot \pi \cdot a \cdot 2b + \frac{1}{2} \cdot \pi \cdot (a+b) \cdot c + a \cdot c = \frac{\pi}{4} \cdot (a \cdot b + a \cdot c + b \cdot c) + a \cdot c$$

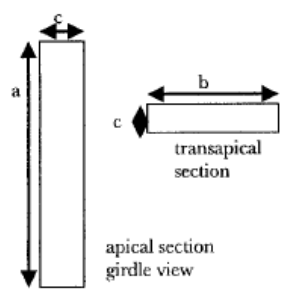
sickle-shaped prism



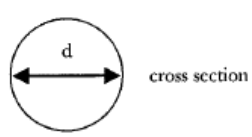
Some diatom genera are formed like a regular prism on a sickle-shaped or lunate base. To calculate this body, a greater half-elliptic prism (dimensions a, b, c) is calculated and a smaller prism is subtracted (the same height, but length (a_2) only from the inner tips of the cell poles and width (b_2) only to the ventral side).

$$V = \frac{1}{4}\pi \cdot a \cdot b \cdot c - \frac{1}{4}\pi \cdot a_2 \cdot b_2 \cdot c = \frac{\pi}{4} \cdot c \cdot (a \cdot b - a_2 \cdot b_2)$$

$$A = \frac{1}{4} \cdot \pi \cdot (a \cdot b - a_2 \cdot b_2) + \frac{1}{4} \cdot \pi \cdot (a+b) \cdot c + \frac{1}{4} \cdot \pi \cdot (a_2 + b_2) \cdot c = \frac{\pi}{4} \cdot ((a \cdot b - a_2 \cdot b_2) + (a+b) \cdot c + (a_2 + b_2) \cdot c)$$



monoraphidioid



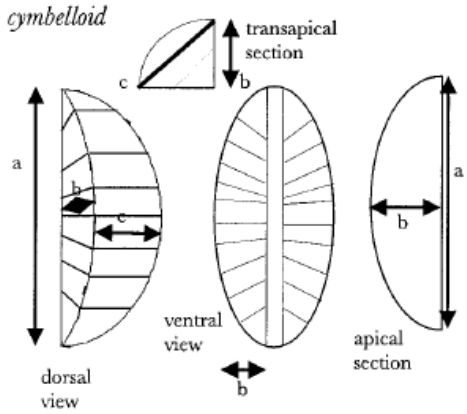
A special case are lunate bodies which are circular in cross-section. The chlorophyte genera *Monoraphidium* and *Kirchneriella* are examples. The maximum diameter of the body is given as d (= $b-b_2$), all other abbreviations as above.

$$V = \frac{d^2}{4} \cdot \left(\frac{(2b - d + a) \cdot \pi^2}{12} + \left(\frac{2b - d + a}{2} \right) \right) = \frac{d^2}{8} \cdot (2b - d + a) \cdot \left(\frac{\pi^2}{6} + 1 \right)$$

GEOMETRIC SHAPES AND EQUATIONS FOR CALCULATION OF BIOVOLUME (cont.)

TABLE 1. Continued.

cymbelloid



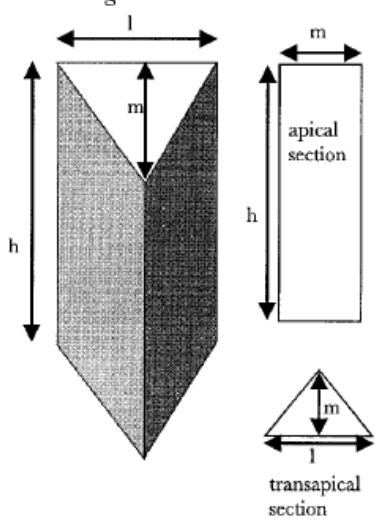
Named after the diatom genus *Cymbella* the body has the shape of a lemon wedge. The volume is calculated as a sector of a prolate spheroid. This ellipsoid is rotating with the transapical axis as radius and with the apical axis as the longer elliptic diameter. c = pervalvar axis on dorsal side; ß = angle between the two transapical sides, to be calculated as

$$\sin\frac{\beta}{2} = \frac{c}{2\cdot b}\ .$$

$$V = \frac{1}{6}\pi\cdot(2b)^2\cdot a\cdot\frac{\beta}{360}$$

$$= \frac{4}{6}\cdot\pi\cdot b^2\cdot a\cdot\frac{\beta}{360}$$

prism on triangle-base



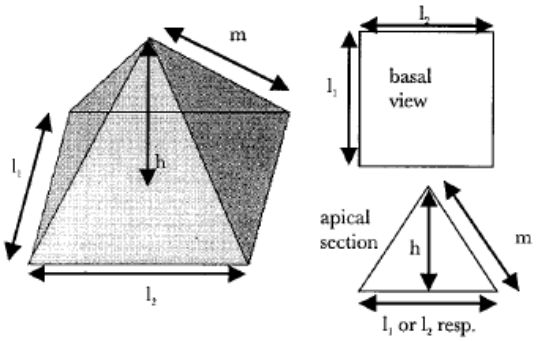
Triangulate diatom species belong mostly to centric genera. In almost every case, the basic triangle is even-sided.

$$V = \frac{1}{2}\cdot l\cdot m\cdot h$$

$$A = 2\cdot 0.5\cdot l\cdot m + 3\cdot l\cdot h$$

$$= l\cdot m + 3\cdot l\cdot h$$

pyramid



In the majority of cells, the basic plate of the pyramid is rectangular. l_1 = length of one side; l_2 = length of another side; m = mantle height on l

$$V = \frac{1}{3}\cdot l_1\cdot l_2\cdot h$$

$$A = l_1\cdot l_2 + 2\cdot\frac{1}{2}\cdot l_1\cdot m$$

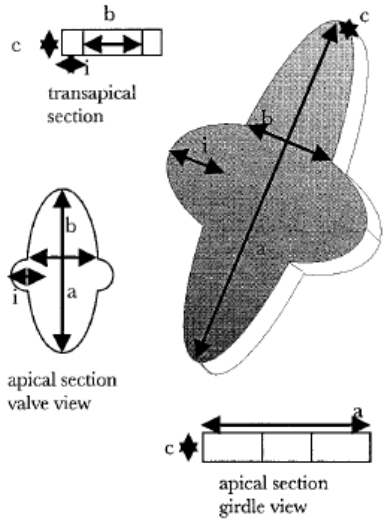
$$+ 2\cdot\frac{1}{2}\cdot l_2\cdot m$$

$$= l_1\cdot l_2 + l_1\cdot m + l_2\cdot m$$

GEOMETRIC SHAPES AND EQUATIONS FOR CALCULATION OF BIOVOLUME (cont.)

TABLE 1. Continued.

elliptic prism with transapical inflations

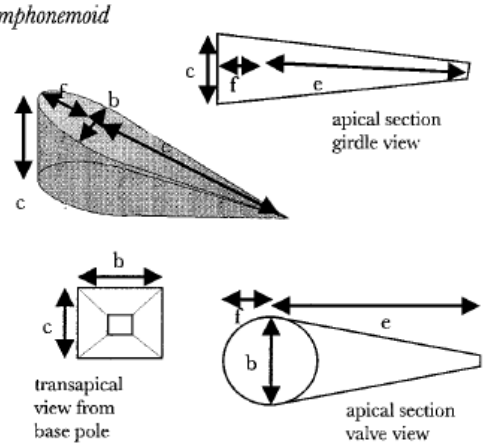


Some pennate diatoms and euglenophytes, e.g. *Tetracyclus* and some *Euglena* sp, are inflated laterally. These inflations can be added to the volume of the central elliptic cylinders by adding two semicircular cylinders (i.e. one cylinder). The surface is measured as two elliptic base plates + two round base plates (= valve view of the inflation) + cylindric mantle (mantle of the inflations) + elliptic mantle (minus 2 plates where the inflations insert). i = diameter of inflation

$$V = \frac{1}{4}\pi \cdot a \cdot b \cdot c + \frac{1}{4}\pi \cdot c \cdot i^2$$
$$= \frac{\pi}{4} \cdot c \cdot (a \cdot b + i^2)$$

$$A = \pi \cdot i \cdot (\frac{1}{2} \cdot i + c) + \frac{1}{2}\pi \cdot (a \cdot b + a \cdot c + b \cdot c) - 2 \cdot c \cdot i$$
$$= \pi \cdot i \cdot (\frac{1}{2} \cdot i + c) + \frac{\pi}{2} \cdot (a \cdot b + a \cdot c + b \cdot c) - 2 \cdot c \cdot i$$

gomphonemoid



Named after the pennate diatom genus *Gomphonema* this body is the most complex in our list because of its double heteropolarity: the two poles of the apical axis are of different width and different height. A very simple but not very accurate approach is a truncated pyramid (Kovala & Larrance 1966). We chose a more realistic model which uses fewer dimensions. The body is a sector of a toroid with a clavate cross-section. This area is calculated as two different half-ellipses. The derivation of the equation is given by Kirschtel (1992). e = length from transapically widest part to base pole; b = maximal transapical width; c = maximal pervalvar height (head pole); f = length from transapically widest part to head pole.

$$V = b \cdot c \cdot \left(\left[\frac{\pi \cdot e}{4} \right] + \left[\frac{f \cdot e}{3} \right] \right)$$

ADDENDUM: Surface areas of complex shapes
For two of the complex bodies in Table 1 we add here equations for the calculation of the surface area. Annotations as in Table 1.

cymbelloid
The surface area of cymbelloids is calculated from the two faces of the body and the dorsal curve.

$$A = \frac{\pi \cdot a \cdot b}{2} + c \cdot \left[b + \frac{\left(\frac{a}{2}\right)^2}{\sqrt{\left(\frac{a}{2}\right)^2 - b^2}} \cdot \sin^{-1}\left(\frac{2 \cdot \sqrt{\left(\frac{a}{2}\right)^2 - b^2}}{a} \right) \right]$$

monoraphidioid
The surface area of this body can be calculated approximately using a double cone, which is assumed to be bent. The mid height of the flexed cone (mh) is calculated from an elliptic circumference.

$$mh = \frac{1}{4} \cdot 2 \cdot \pi \cdot \sqrt{\frac{\left(\frac{a}{2}\right)^2 + \left(b - \frac{d}{2}\right)^2}{2}} = \frac{\pi}{2} \cdot \sqrt{\frac{\left(\frac{a}{2}\right)^2 + \left(b - \frac{d}{2}\right)^2}{2}}$$

$$A = 2 \cdot \pi \cdot \frac{d}{2} \cdot mh = \pi \cdot d \cdot mh$$

MODELS FOR BIOVOLUME CALCULATIONS OF MICROALGAL TAXA

TABLE 2. Application of geometric models for biovolume calculations of microalgal taxa. The table is sorted according to higher taxonomic groups and lists the genera alphabetically. The annotations (A) are given at the end of the table according to their numbers. We do not claim completeness for the genera or for the annotations. There may be significant deviations from this generic approach. The applied shapes should be checked carefully.

Genus	Shape	A
1. Cyanobacteria		
Agmenellum De Breb.	sphere	
Ammatoidea W & GS West	sphere	
Anabaena Bory	sphere	1,2
Anabaenopsis (Wol.) V.Mill.	cylinder	
Anacystis Menegh.	prolate spheroid	
Aphanizomenon Morr.	cylinder	
Aphanocapsa Naeg.	sphere	
Aphanothece Naeg.	prolate spheroid	
Arthronema Kom. & Lukavsky	cylinder	
Arthrospira Stizenb.	cylinder	
Aspalatia Erceg.	prolate spheroid	
Aulosira Kirchner	cylinder	
Bacularia Borzi	prolate spheroid	
Blennothrix Kütz.	cylinder	
Borzia Cohn	cylinder	
Calothrix Ag.	cylinder	3
Camptothrix W & GS West	cylinder	
Camptylonemopsis Desikchary	cylinder	
Catella Alvik	prolate spheroid	
Chamaecalyx Kom. & Anagnostidis	sphere	
Chlorogloea Wille	prolate spheroid	
Chroococcidiopsis Geitler	sphere	
Chroococcidium Geitler	sphere	
Chroococcopsis Geitler	prolate spheroid	
Chroococcus Naeg.	sphere	
Chroostipes Pasch.	prolate spheroid	
Clastidium Kirchner	cylinder	
Coccopedia Troick.	prolate spheroid	
Coelosphaerium Naeg.	prolate spheroid	
Coleodesmium Borzi	cylinder	
Coleodesmiumopsis Dutt	cylinder	
Crinalium Crow	cylinder	
Cyanarcus Pascher	cylinder	
Cyanobacterium Rippka & Coh.-Baz.	prolate spheroid	
Cyanobium Rippka & Cohen-Bazire	prolate spheroid	
Cyanocatena Hind.	prolate spheroid	
Cynocystis Borzi	sphere	
Cyanodermatium Geitler	prolate spheroid	
Cyanodictyon Pasc.	prolate spheroid	
Cyanogranis Hind.	prolate spheroid	
Cyanokybus Schill.	sphere	
Cyanonephron Hickel	prolate spheroid	
Cyanophanon Geit.	cylinder	
Cyanosaccus Lukas & Golub.	prolate spheroid	
Cyanostylon Geitler	prolate spheroid	
Cyanothece Kom.	prolate spheroid	
Cylindrospermopsis Seen. & S.Raju	cylinder	
Cylindrospermum Kütz.	cylinder	
Dermocarpella Lemm.	sphere	
Dichothrix Zanard.	cylinder	
Dzensia Voronich.	prolate spheroid	
Enthophysalis Kütz.	prolate spheroid	
Epilithia Erceg.	prolate spheroid	
Erceg.ia De Toni	prolate spheroid	
Eucapsa Clem. & Sh.	prolate spheroid	
Fortiea De Toni	cylinder	
Gardnerula De Toni	cylinder	
Geitleribactron Kom.	cylinder	
Gloeobacter Rippka	prolate spheroid	
Gloeocapsa Kütz.	sphere	
Gloeocapsopsis Geitler	sphere	
Gloeothece Naeg.	prolate spheroid	
Gloeotricha J Ag.	cylinder	3
Gomontiella Teodor.	cylinder	
Gomphosphaeria Kütz.	sphere	2
Hassallia Berk.	cylinder	
Heteroleibleinia (Geitler) Hoffm.	cylinder	
Homoeothrix (Thur.) Kirchner	cylinder	
Hormathonema Erceg.	prolate spheroid	
Hormoscilla Anagn. & Kom.	cylinder	
Hydrococcus Kütz.	prolate spheroid	
Hydrocoleum Kütz.	cylinder	
Hydrocoryne Schw.	cylinder	
Hyella Born.&Flah.	cylinder	
Isactis Thuret	cylinder	3
Isocystis Borzi	cylinder	
Jaaginema Anagn. & Kom.	cylinder	
Johannesbaptistia De Toni	prolate spheroid	
Katagnymene Lemmermann	cylinder	
Komvophoron Anagn. & Kom.	cylinder	
Kyrtuthrix Erceg.	cylinder	
Leibleinia Hoffm.	cylinder	
Lemmermanniella Geitler	prolate spheroid	
Leptolyngbya Anagn. & Kom.	cylinder	
Limnothrix Meffert	cylinder	
Lithocapsa Erceg.	prolate spheroid	
Lithodiscus Erceg.	cylinder	
Lyngbya J.G. Ag	cylinder	
Lyngbyopsis Gardn.	cylinder	
Merismopedia Meyen	cube	
Microchaete Thuret	cylinder	3
Microcoleus Desm.	cylinder	
Microcrocis Richter	prolate spheroid	
Microcystis Kütz.	sphere	
Myxobaktron Schmidle	prolate spheroid	
Myxohyella Geitler	prolate spheroid	
Myxosarcina Printz	prolate spheroid	
Nematoradaisia Geitler	prolate spheroid	
Nodularia Mert.	cylinder	
Nostoc Vaucher	cylinder	
Onkonema Geitler	prolate spheroid	
Oscillatoria Vaucher	cylinder	
Palikiella Claus	cylinder	
Paracapsa Naum.	prolate spheroid	
Pascherinema De Toni	cylinder	
Petalonema Berk.	cylinder	
Phormidium Kütz.	cylinder	
Pilgeria Schmidle	prolate spheroid	
Placoma Schousboe	prolate spheroid	
Planktolyngbya Anagn. & Kom.	cylinder	
Planktothrix Anagn. & Kom.	cylinder	
Plectonema Thuret	cylinder	
Pleurocapsa Thuret	cylinder	
Podocapsa Erceg.	prolate spheroid	
Porphyrosiphon Kütz.	cylinder	
Proterendothrix W & GS West	cylinder	

MODELS FOR BIOVOLUME CALCULATIONS OF MICROALGAL TAXA (cont.)

TABLE 2. Continued.

Genus	Shape	A
Pseudanabaena Lauterbach	cylinder	
Pseudocapsa Erceg.	sphere	
Pseudoncobyrsa Geitler	prolate spheroid	
Pseudophormidium (Forti) Anagn.&Kom.	cylinder	
Pseudoscytonema Elenk.	cylinder	
Radaisia Sauv.	cylinder	
Radaisiella Geitler	cylinder	
Radiocystis Skuja	sphere	
Raphidiopsis Fritsch & Rich	cylinder	
Richelia Schmidt	cylinder	
Rivularia (Roth) Ag.	cylinder	3
Rhabdoderma Schmidle	prolate spheroid	
Rhabdogloea Schr.	prolate spheroid	
Rhodostichon Geitler & Pascher	prolate spheroid	
Romeria Koczw.	cylinder	
Sacconema Borzi	cylinder	
Schizothrix Kütz.	cylinder	
Scytonema Ag.	cylinder	
Scytonemopsis Kis.	cylinder	
Sinaiella Gruia	cylinder	
Siphonema Geitler	cylinder	
Sirocoleum Kütz.	cylinder	
Sokolovia Elenk.	cylinder	
Solentia Erceg.	prolate spheroid	
Spirulina Turpin	cylinder	
Stanieria Komarek & Anagnostidis	sphere	
Starria Lang	cylinder	
Stichosiphon Geitler	cylinder	
Symploca Kütz.	cylinder	
Synechococcus Näg.	sphere	
Synechocystis Sauv.	sphere	
Tetrarcus Skuja	prolate spheroid	
Tolypothrix Kütz.	cylinder	
Trichodesmium Ehr.	cylinder	
Trichormus Kom. & Anagn.	cylinder	
Tubiella Hollerb.	prolate spheroid	
Tychonema Anagn. & Kom.	cylinder	
Wollea Born.& Flah.	cylinder	
Wolskyella Claus	prolate spheroid	
Xenococcus Thuret	prolate spheroid	
2. Chrysophyceae		
Actinomonas Kent	sphere	
Amphichrysis Kors.	prolate spheroid	
Apedinella Throng.	prolate spheroid	
Aureococcus Hargr. & Sieburth	sphere	
Calycomonas Loh.	cone	
Chromulina Cienk.	sphere	
Chrysapsis Pascher	prolate spheroid	
Chrysocapsa Pascher	sphere	
Chrysococcus Klebs	prolate spheroid	
Chrysopora Pascher	prolate spheroid	
Chrysosaccus Bour.	prolate spheroid	
Chrysosphaera (Pascher) Bour.	sphere	
Chrysosphaerella Lauterborn	prolate spheroid	
Chrysotilos Pascher	prolate spheroid	
Ciliophrys Cienk.	sphere	
Dictyocha Ehr.	sphere	
Dinobryon Ehr.	prolate spheroid	1,4
Epicystis Pascher	sphere	
Epipyxis Ehr.	ellipsoid	
Gloeochrysis Pascher	prolate spheroid	
Hydrurus Ag.	ellipsoid	
Kephyrion Pascher	prolate spheroid	
Mallomonas Perty	prolate spheroid	
Mallomonopsis Matvienko	prolate spheroid	
Meringosphaera Lohmann	sphere	
Mesocena Ehr.	sphere	
Microglena Ehr.	prolate spheroid	
Monochrysis Skuja	prolate spheroid	
Ochromonas Wyss.	cone+half sphere	
Octatis Schiller	sphere	
Parapedinella Peders. & Thomsen	sphere	
Paraphysomonas de Saedeleer	sphere	5
Pedinella Wyssotzky	sphere	
Pelagococcus Norris	sphere	
Phaeaster (Scheffel) Bourelly	ellipsoid	
Phaeobotrys Ettl	sphere	
Pseudokephyrion (Pascher) Schm.	ellipsoid	
Pseudopedinella Carter	truncated cone	
Pyramidochrysis Pascher	cone	
Saccochrysis Kors.	prolate spheroid	
Sarcinochrysis Geit.	prolate spheroid	
Sphaerapsis Schiller	sphere	
Sphaleromantis Pascher	cone	
Spumella Cienk.	cone+half sphere	
Syncrypta Ehr.	ellipsoid	
Synura Ehr.	prolate spheroid	
Tetrasporopsis Lemmermann	sphere	
Uroglena Ehr.	cone+half sphere	5
3. Xanthophyceae		
Akanthochloris Pas.	sphere	
Arachnochloris Pascher	sphere	
Botryochloris Pascher	sphere	
Botryodiopsis Ettl	sphere	
Brachynema Alvik	cylinder	
Bumilleria Borzi	cylinder	
Bumilleriopsis Printz	prolate spheroid	
Centritactus Lemm.	prolate spheroid	
Charciopsis Borzi	ellipsoid	
Chlorallantes Pasch.	ellipsoid	
Chlorobotrys Bohlin	prolate spheroid	
Chlorochloster Pascher	2 cones	
Chlorothecium Borzi emend. Pascher	ellipsoid	
Diachros Pascher	sphere	
Ellipsoidion Pascher	ellipsoid	
Gloeobotrys Pascher	prolate spheroid	
Gloeopodium Pascher	ellipsoid	
Goniochloris Geitler	prism on triangle	
Heterothrix Pascher	cylinder	
Heterotrichella Reis.	cylinder	
Lutherella Pascher	sphere	
Meringosphaeria Loh. em. Pascher	sphere	
Monallantes Pasch.	prolate spheroid	
Monodus Chodat	cone+half sphere	
Neonema Pascher	cylinder	
Nephrodiella Pascher	prolate spheroid	
Ophiocytium Naeg.	cylinder	
Pleurochloris Pascher	sphere	5
Pseudostaurastrum Chodat	4 boxes	
Pseudotetraëdon Pascher	box	
Schilleriella Pascher	cylinder + half sphere	
Tetraëdiella Pascher	prism on triangle	
Trachychloron Pascher	ellipsoid	
Trachydiscus Ettl	ellipsoid	
Tribonema Derbes & Solier	cylinder	
Vischeria Pascher	sphere	
4. Bacillariophyceae		
Acanthoceras Honig.	elliptic prism	
Achnanthes Bory	elliptic prism	6-9

MODELS FOR BIOVOLUME CALCULATIONS OF MICROALGAL TAXA (cont.)

TABLE 2. Continued.

Genus	Shape	A
Achnanthidium Kütz.	elliptic prism	6-9
Actinella FW Lewis	box + half-elliptic prism	
Actinocyclus Ehr.	cylinder	
Actinoptychus Ehr.	cylinder	
Adoneis Andr.& Riv.	prism on parallelogram	
Amphipleura Kütz.	elliptic prism	
Amphiprora Ehr.	elliptic prism	
Amphitetras Ehr.	box	
Amphora Ehr.	cymbelloid	10
Anaulus Ehr.	elliptic prism	
Aneumastus Mann & Stickle	elliptic prism	
Anomoeoneis Pfitzer	elliptic prism	
Anorthoneis Grun.	elliptic prism	
Arachnoidiscus Deane	cylinder	
Arcocellulus Hasle et al.	elliptic prism	6
Ardissonea De Not.	box	
Asterionella Hassall	box + 2 cylinders	
Asterionellopsis Round	prism on triangle	
Asterolampra Ehr.	cylinder	
Asteromphalus Ehr.	cylinder	11
Attheya T West	elliptic prism	
Aulacodiscus Ehr.	cylinder	
Aulacoseira Thwait.	cylinder	
Auliscus Ehr.	elliptic prism	
Auricula Castr.	cymbelloid	
Azpeitia M Perag.	cylinder	
Bacillaria Gmelin	box	
Bacteriastrum Shad.	cylinder	
Bacterosira Gran	cylinder	
Banquisia Paddock	box	
Bellerochea Van Heurck	prism on triangle	
Berkeleya Greville	elliptic prism	8
Biddulphia Gray	elliptic prism	12
Biddulphiopsis Von Stosch & Sim.	elliptic prism	
Biremis Mann & Cox	elliptic prism	8
Bleakeleya Round	box + elliptic prism	13
Brachysira Kütz.	elliptic prism	
Brebissonia Grun.	elliptic prism	
Brockmanniella Hasle et al.	elliptic prism	
Caloneis Cleve	elliptic prism	8
Campylodiscus Ehr.	elliptic prism	6
Campyloneis Grun.	elliptic prism	
Campylopyxis Medlin	elliptic prism	
Campylosira Grun.	half-elliptic prism	6
Catacombas Williams & Round	box	
Catenula Mereschk.	half-elliptic prism	
Cavinula Mann & Stickle	elliptic prism	
Centronella Voigt	3 boxes	
Cerataulina H.Perag.	cylinder	
Cerataulus Ehr.	elliptic prism	
Chaetoceros Ehr.	elliptic prism	12, 14
Chrysanthemodiscus A.Mann	cylinder + 2 half spheres	
Climacodium Grun.	elliptic prism + 4 cones	12
Climaconeis Grun.	box	
Climacosphenia Ehr.	box + elliptic prism	
Cocconeis Ehr.	elliptic prism	
Corethron Castr.	cylinder + 2 half spheres	
Coscinodiscus Ehr.	cylinder	15
Cosmioneis Mann & Stickle	elliptic prism	
Craticula Grun.	elliptic prism	
Ctenophora Williams & Round	elliptic prism	8
Cuneolus Giffen	gomphonemoid	11
Cyclophora Castr.	elliptic prism	
Cyclostephanos Round	cylinder	
Cyclotella (Kütz.) De Breb.	cylinder	
Cylindrotheca Rabenh.	prolate spheroid + 2 cylinders	16
Cymatopleura W Smith	box	17
Cymatoneis Cleve	prism on parallelogram	
Cymatosira Grun.	prism on parallelogram	
Cymbella CA Ag.	cymbelloid	18
Cymbellonitzschia Hustedt	half-elliptic prism	
Dactyliosolen Castr.	cylinder	
Delphineis Andrews	elliptic prism	
Denticula Kütz.	elliptic prism	8
Detonula Schütt	cylinder	
Diadesmis Kütz.	elliptic prism	
Diatoma De Cand.	elliptic prism	8-9
Diatomella Greville	elliptic prism	
Dictyoneis Cleve	elliptic prism	17
Didymosphenia M Schmidt	elliptic prism	17
Dimeregramma Ralfs	elliptic prism	7
Dimeregrammopsis Ricard	elliptic prism	
Diplomenora Blazé	elliptic prism	
Diploneis Ehr.	elliptic prism	17
Ditylum JW Bailey	prism on triangle	
Donkinia Ralfs	prism on parallelogram	
Druridgia Donkin	elliptic prism	
Ellerbeckia Crawford	cylinder	
Encyonema Kütz.	cymbelloid	
Endictya Ehr.	cylinder	
Entomoneis Ehr.	elliptic prism	11
Ephemera Paddock	box	
Epithemia De Breb.	cymbelloid	18
Ethmodiscus Castr.	cylinder	
Eucampia Ehr.	elliptic prism	11
Eucocconeis Cleve	elliptic prism	
Eunotia Ehr.	sickle-shaped prism	19
Eunotogramma Weise	half-elliptic prism	
Eupodiscus Ehr.	cylinder	
Extubocellus Hasle et al.	elliptic prism	
Falcula Voigt	sickle-shaped prism	19
Fallacia Stickle & Mann	elliptic prism	
Fragilaria Lyngbye	elliptic prism	7,8, 20
Fragilariforma Williams & Round	elliptic prism	
Fragilariopsis Hust.	elliptic prism	
Frickea Heiden	box	
Frustulia Rabenh.	elliptic prism	
Gephyria Arnott	elliptic prism	6
Glyphodesmis Grev.	elliptic prism	
Gomphocymbella O Müller	cymbelloid	
Gomphoneis Cleve	elliptic prism	21
Gomphonema CA Ag.	gomphonemoid	18
Gomphonemopsis Medlin	gomphonemoid	
Gomphonitzschia Grun.	gomphonemoid	
Gomphoseptatum Medlin	gomphonemoid	
Gomphotheca Hendey & Sims	box	
Gonoiceros Perag.	elliptic prism	
Gossleriella Schütt	cylinder	
Grammatophora Ehr.	elliptic prism	8
Guinardia H Perag.	cylinder	
Gyrosigma Hassall	prism on parallelogram	
Hannaea Patrick	sickle-shaped prism	
Hantzschia Grun.	box	
Haslea Simonsen	elliptic prism	
Helicotheca Ricard	elliptic prism	
Hemiaulus Heiberg	elliptic prism	
Hemidiscus Wallich	cymbelloid	
Hustedtiella Simon.	elliptic prism	
Hyalodiscus Ehr.	sphere	22

MODELS FOR BIOVOLUME CALCULATIONS OF MICROALGAL TAXA (cont.)

TABLE 2. Continued.

Genus	Shape	A
Hyalosynedra Williams & Round	box	
Hydrosera Wallich	prism on triangle	23
Hydrosilicon Brun	box	17
Hydrosira Mills	cylinder	
Isthmia CA Ag.	elliptic prism	
Lampriscus A Schm.	prism on triangle	
Lauderia Cleve	cylinder	
Lennoxia Thomsen & Buck	half-elliptic prism	7
Leptocylindrus Cleve	cylinder	
Leyanella Hasle et al.	elliptic prism	6
Licmophora CA Ag.	gomphonemoid	
Lioloma Hasle	box	
Lithodesmium Ehr.	prism on triangle	
Lunella Snoeijs	half-elliptic prism	
Luticola Mann	elliptic prism	
Lyrella Karajeva	elliptic prism	
Manguinea Paddock	box	
Martyana Round	elliptic prism	21
Mastogloia Thw.	elliptic prism	
Melosira CA Ag.	cylinder	24
Membraneis Padd.	elliptic prism	
Meridion CA Ag.	gomphonemoid	
Microtabella Round	elliptic prism	
Minidiscus Hasle	cylinder	
Minutocellus Hasle et al.	elliptic prism	6
Nanoneis Norris	box	
Navicula Bory	elliptic prism	7,8
Neidium Pfitzer	elliptic prism	8
Neodelphineis Takano	box	
Neodenticula Akiba & Yanagisawa	elliptic prism	
Neostreptotheca von Stosch	box	
Neosynedra Williams & Round	box	
Nitzschia Hassall	prism on parallelogram	25
Odontella CA Ag.	elliptic prism	12
Opephora Petit	elliptic prism	21
Orthoseira Thwaites	cylinder	
Oxyneis Round	elliptic prism	17
Palmeria Greville	cymbelloid	
Papiliocellulus Hasle et al.	elliptic prism	
Paralia Heiberg	cylinder	
Parlibellus Cox	elliptic prism	
Perissonoë Andrews & Stoelzel	box	
Peronia De Breb. & Arnott	gomphonemoid	
Petrodyction Mann	elliptic prism	
Petroneis Stickle & Mann	box	26
Phaeodactylum Boh.	half-elliptic prism	27
Pinnularia Ehr.	box	9
Placoneis Mereschk.	elliptic prism	7
Plagiodiscus Jurilj	half-elliptic prism	
Plagiogramma Grev.	elliptic prism	
Plagiogrammopsis Hasle et al.	elliptic prism	8
Plagiotropis Pfitzer	elliptic prism	8
Planktoniella Schütt	cylinder	28
Pleurosigma W Sm.	prism on parallelogram	
Pleurosira Trevisan	elliptic prism	
Podocystis Bailey	elliptic prism	
Podosira Ehr.	cylinder + 2 half spheres	29
Porosira Jörgensen	cylinder	
Proboscia Sundstr.	cylinder	
Progonoia Schrader	elliptic prism	17
Proschkinia Karay.	elliptic prism	
Psammodictyon Mann	elliptic prism	17
Psammodiscus Round & Mann	cylinder	
Pseudogomphone-ma Medlin	gomphonemoid	
Pseudoguinardia Von Stosch	cylinder	
Pseudohimantidium Hust. & Krasske	cymbelloid	
Pseudonitzschia H Perag.	prism on parallelogram	8
Pseudosolenia Sundström	cylinder	
Pseudostaurosira Williams & Round	box	20
Pseudotriceratium Grun.	prism on triangle	
Pteroncola Holmes & Croll	elliptic prism	
Punctastriata Williams & Round	elliptic prism	
Reimeria Kociolek & Stoermer	elliptic prism	
Rhabdonema Kütz.	box	
Rhaphoneis Ehr.	prism on parallelogram	
Rhizosolenia Ehr.	cylinder	
Rhoicosphenia Gru.	gomphonemoid	
Rhoikoneis Grun.	elliptic prism	6
Rhopalodia O Müller	cymbelloid	
Roperia Grun.	elliptic prism	
Rutilaria Greville	elliptic prism	
Sceptroneis Ehr.	box	
Scolioneis Mann	gomphonemoid	
Scoliopleura Grun.	box	
Scoliotropis Cleve	box	
Sellaphora Meresch.	box	
Seminavis Mann	cymbelloid	
Semiorbis Patrick	sickle-shaped prism	
Sheshukovia Glezer	prism on triangle	
Skeletonema Grev.	cylinder + 2 half spheres	30
Stauroneis Ehr.	elliptic prism	8
Staurophora Mereschkowsky	elliptic prism	
Stauropsis Meunier	elliptic prism	
Staurosira (Ehr.) Williams & Round	elliptic prism	
Staurosirella Williams & Round	elliptic prism	20
Stellarima Hasle & Sims	cylinder	
Stenoneis Cleve	box	
Stenopterobia De Breb.	prism on parallelogram	
Stephanodiscus Ehr.	cylinder	
Stephanopyxis (Ehr.) Ehr.	cylinder + 2 half spheres	
Stictocyclus A Mann	cylinder	
Stictodiscus Greville	cylinder	31
Streptotheca Shrubs.	box	
Striatella CA Ag.	elliptic prism	8
Subsilicea Von Stosch & Reim.	box	
Surirella Turpin	elliptic prism	
Synedra Ehr.	box	26
Synedropsis Hasle et al.	elliptic prism	
Synedrosphenia (H Perag.) Zarag.	gomphonemoid	
Tabellaria Ehr.	box	17
Tabularia Williams & Round	elliptic prism	8
Terpsinoë Ehr.	box + 6 half cylinders	
Tetracyclus Ralfs	elliptic prism with inflations	
Thalassioneis Round	elliptic prism	
Thalassionema Gru.	box	
Thalassiophysa Conger	elliptic prism	11
Thalassiosira Cleve	cylinder	
Thalassiothrix Cleve & Grun.	box	
Toxarium JW Bail.	2 boxs + elliptic prism	30
Toxonidea Donkin	half-elliptic prism	
Trachyneis Cleve	elliptic prism	
Trachysphenia Petit	elliptic prism	
Triceratium Ehr.	prism on triangle	
Trichotoxon Reid & Round	box	
Trigonium Cleve	prism on triangle	

MODELS FOR BIOVOLUME CALCULATIONS OF MICROALGAL TAXA (cont.)

TABLE 2. Continued.

Genus	Shape	A
Tropidoneis Cleve		
Tryblionella W Smith	elliptic prism	8
Undatella Paddock & Sims	half-elliptic prism	17
Urosolenia Round & Crawford	cylinder	
5.Raphidophyceae		
Chattonella Biech.	cone+half sphere	
Fibrocapsa Toriumi & Takano	prolate spheroid	
Heterosigma Hada	truncated cone & halfsphere	
Olisthidscus Carter	prolate spheroid	
Oltmannsia Schiller	prolate spheroid	
6.Prymnesiophyceae		
Acanthoica Loh.	prolate spheroid	
Algirosphaera Schlauder	prolate spheroid	
Alisphaera Heimdal	sphere	
Anaplosolenia Defl.	2 cones	
Anthosphaera Kampt.	sphere	
Balaniger Thomsen & Oates	prolate spheroid	
Braarudosphaera Defl.	sphere	
Calcidiscus Kampt.	sphere	2
Calciopappus Gaar. & Ramsfjell	cone	
Calciosolenia Gran	cylinder	
Calyptrolithina Heimdal	prolate spheroid	
Calyptrolithophora Heimdal	prolate spheroid	
Calyptrosphaera Loh.	sphere	
Caneosphaera Gaarder	sphere	
Ceratolithus Kampt.	sphere	
Chrysidalis Schiller	cylinder	
Chrysochromulina Lackey	prolate spheroid	1,2
Coccolithus Schwarz	sphere	
Corisphaera Kampt.	sphere	
Coronosphaera Kamptner	sphere	
Corymbellus Green	prolate spheroid	
Crenalithus Roth	sphere	
Cricosphaera Braarud	sphere	5
Cruciplacolithus Hay & Mohler	sphere	
Crystallolithus Gaard.& Mark.	sphere	
Daktylethra Gartner	prolate spheroid	
Derepyxis Stokes	ellipsoid	
Deutschlandia Loh.	sphere	
Diacromena Prauser	prolate spheroid	
Dicrateria Parke	sphere	
Discolithina Loebl. & Tappan	sphere	
Discosphaera Haeckel	sphere	
Emiliana Hay & Moh.	cylinder	
Florisphaera Okada & Honjo	prolate spheroid	
Gephyrocapsa Kampt.	sphere	
Gliscolithus Norris	sphere	
Halopappus Loh.	prolate spheroid	33
Helicosphaera Kampt.	sphere	
Helladosphaera Kampt.	sphere	
Homozygosphaera Defl.	sphere	
Hymenomonas Stein	prolate spheroid	
Imantonia Reynolds	sphere	
Isochrysis Parke	prolate spheroid	
Michaelsarsia Gran	sphere	5
Oolithotus Reinh.	sphere	
Ophiaster Gran em. Mant. & Oakes	prolate spheroid	
Palusphaera Lecal emend Norris	sphere	
Pappomonas Mant. & Oakes	sphere	
Papposphaera Tangen	sphere	
Parachrysidalis Hulburt	prolate spheroid	
Pavlova Butcher	prolate spheroid	
Periphyllophora Kamp.	prolate spheroid	
Phaeocystis Lagerh.	sphere	
Platychrysis Geitler	cylinder	
Pleurochrysis Pring. em. Gayr. & Fr.	sphere	
Pontosphaera Loh.	sphere	
Pymnesium Mass. ex Conrad	cone+half sphere	
Rhabdosphaera Haeckel	sphere	
Scyphosphaera Loh.	sphere	
Sphaerocalyptra Defl.	sphere	
Syracolithus (Kampt.) Defl.	sphere	
Syracosphaera Loh. em Gaarder	sphere	
Tesselaria Playfair	ellipsoid	
Thorosphaera Ostf.	sphere	
Umbellosphaera Paasche	sphere	
Umbilicosphaera Loh.	sphere	
Zygosphaera Kampt.	sphere	
7. Cryptophyceae		
Chilomonas Ehr.	cone+half sphere	34
Chroomonas Hansg.	cone+half sphere	34
Cryptomonas Ehr.	prolate spheroid	1
Geminigera Hill	prolate spheroid	
Goniomonas Stein	cone+half sphere	
Hemiselmis Parke	prolate spheroid	
Hillea Schiller	prolate spheroid	
Isoselmis Butcher	cone+half sphere	
Leucocryptos Butch.	cone+half sphere	
Plagioselmis Butch.	cone+half sphere	
Rhinomonas Hill & Wetherbee	cone+half sphere	
Rhodomonas Karst.	cone+half sphere	1
Teleaulax Hill	cone+half sphere	
8. Dinophyceae		
Acanthogonyaulax (Kof.) Graham	ellipsoid	
Adenoides Balech	elliptic prism	
Alexandrium Halim	ellipsoid	35
Amphidiniopsis Wol.	elliptic prism	
Amphidinium Clap. & Lachmann	ellipsoid	36
Amphidoma Stein	2 cones	
Amphisolenia Stein	cylinder	
Amylax Meunier	cone+half sphere	
Balechina Loebl. Jr. & Loebl. III	prolate spheroid	
Bernardinium Chod.	ellipsoid	
Blepharocysta Ehr.	sphere	
Boreadinium Dodge & Hermes	prolate spheroid	
Brachydinium Tayl.	cylinder + 4 cones	
Centrodinium Kof.	2 cones	
Ceratium Schrank	ellipsoid + 2 cones + cylinder	1, 37
Ceratocorys Stein	half sphere	
Citharistes Stein	ellipsoid	
Cladopyxis Stein	sphere	
Cochlodinium Schütt	prolate spheroid	
Coolia Meunier	ellipsoid	
Corythodinium Loeb Jr. & Loebl. III	2 cones	
Crypthecodinium Biecheler	prolate spheroid	
Cymbodinium Cach. & Cach.	sphere	
Cystodinedria Pascher	ellipsoid	

MODELS FOR BIOVOLUME CALCULATIONS OF MICROALGAL TAXA (cont.)

TABLE 2. Continued.

Genus	Shape	A
Cystodinium Klebs	prolate spheroid	
Desmonastrix Pascher	prolate spheroid	
Dinococcus (Wol.) Fott	pyramid on triangular base	
Dinophysis Ehr.	ellipsoid	38
Dinosphaera Kof. & Michener	ellipsoid	
Diplopelta Stein	ellipsoid	
Diplopsalis Bergh	cone+half sphere	
Diplopsalopsis Meunier	sphere	
Ensiculifera Balech	cone+half sphere	
Erythropsidinium Silva	ellipsoid	
Fragilidium Balech	sphere	
Gambierdiscus Adachi & Fuk.	ellipsoid	
Glenodiniopsis Wol.	ellipsoid	
Glenodinium Ehr.	ellipsoid	
Goniodoma Stein	sphere	
Gonyaulax Diesing	2 cones	39
Gotoius Abe	prolate spheroid	
Gymnodinium Stein	ellipsoid	40
Gyrodinium Kof. & Swezy	ellipsoid	41
Hemidinium Stein	ellipsoid	
Herdmania Dodge	prolate spheroid	
Heterocapsa Stein	2 cones	
Heterodinium Kof.	2 cones	
Histioneis Stein	ellipsoid	
Katodinium Fott	cylinder + cone	42
Kofoinium Pavillard	sphere	
Leptodiscus Hertwig	cylinder	
Lingulodinium Dodge	ellipsoid	
Lissodinium Matz. em.Car.-Moore	ellipsoid	
Lophodinium Lemm.	2 cones	
Mesoporos Lillick	elliptic prism	
Micranthodinium	sphere	
Nematodinium Kof. & Swezy	ellipsoid	
Noctiluca Suriray	sphere	
Oblea Balech	sphere	
Ornithocercus Stein	half sphere	43
Oodinium Chatton	ellipsoid	
Ostreopsis Schmidt	ellipsoid	
Oxophysis Kof.	2 cones	
Oxyrrhis Dujardin	prolate spheroid	
Oxytoxum Stein	prolate spheroid	
Palaeophalacroma Schiller	prolate spheroid	
Pentapharsodinium Indel. & Loebl.	ellipsoid	
Peridiniella Kof. & Mich. em. Bal.	ellipsoid	
Peridinium Ehr.	ellipsoid	44
Peridinopsis Lemm.	ellipsoid	
Phalacroma Stein	ellipsoid	
Pheopolykrikos (Chatton) Mat.&Fuk.	ellipsoid	
Pleromonas Pascher	prolate spheroid	
Podolampas Stein	cone	
Polykrikos Bütschli	ellipsoid	
Preperidinium Mangin	ellipsoid	
Promatodinium Cach. & Cach.-Enj.	sphere	
Pronoctiluca Fabre-Domergue	cone+half sphere	
Prorocentrum Ehr.	ellipsoid	45
Protaspis Skuja	ellipsoid	
Protoceratium Bergh	sphere	
Protoperidinium Bergh	2 cones	46
Pseudactiniscus Bursa	sphere	
Ptychodiscus Stein	prolate spheroid	
Pyrocystis Murray	ellipsoid	
Pyrodinium Plate	ellipsoid	
Pyrophacus Stein	ellipsoid	
Roscoffia Balech	ellipsoid	
Schuetiella Balech	2 cones	
Scrippsiella Balech	ellipsoid	
Sinophysis Nie & Wang	ellipsoid	
Spatulodinium Cach. & Cach.	ellipsoid	
Sphaerodinium Woloszynska	ellipsoid	
Spiraulax Kof.	2 cones	
Spiromonas Dujardin	cone+half sphere	
Stylodinium Klebs	sphere	
Symbiodinium Freudenthal	prolate spheroid	
Tetradinium Klebs	pyramid	
Thecadinium Kof. & Skogsberg	ellipsoid	
Thompsodinium Bour.	ellipsoid	
Torodinium Kof. & Swezy	prolate spheroid	
Triposolenia Kof.	cone + 2 cylinders	47
Warnowia Lindem.	prolate spheroid	
Woloszynskia Thompson	ellipsoid	
9. Euglenophyceae		
Colacium Ehr.	ellipsoid	
Euglena Ehr.	half ellipsoid + cone on ellitpic base	1, 34
Eutreptia Perty	cylinder + cone	
Eutreptiella De Cunha	cylinder + cone	
Lepocinclis Perty	2 cones	5
Phacus Dujardin	elliptic prism	34
Trachellomonas Ehr. emend. Defl.	ellipsoid	
10. Chlorophyta		
10a. Prasinophyceae		
Bathyococcus Eikr. & Throndsen	sphere	
Cymbomonas Schill.	prolate spheroid	
Halosphaera Schmitz	sphere	5
Mamiella Moestrup	prolate spheroid	
Mantoniella Desik.	prolate spheroid	
Micromonas Mant. & Parke	prolate spheroid	
Monomastix Scheffel	prolate spheroid	
Nephroselmis Stein	sphere	5
Pachysphaera Ostf.	prolate spheroid	
Pedinomonas Kors.	prolate spheroid	
Polyblepharides Dang.	cone	
Prasinocladus Kuck.	prolate spheroid	
Pseudoscourfielda Manton	ellipsoid	
Pterosperma Pouc.	sphere	
Pycnococcus Guill.	sphere	
Pyramimonas Schm.	cone	
Resultor Moestrup	sphere	5
Scourfielda GS West	ellipsoid	
Tetraselmis Stein	prolate spheroid	
10b. Chlorophyceae		
Actinastrum Lagerh.	cylinder+2 cones	48
Actinochloris Kors.	prolate spheroid	
Amphikrikos Kors.	ellipsoid	
Ankistrodesmus Corda	cylinder+2 cones	49
Ankyra Fott	2 cones	
Apodochloris Kom.	cone+half sphere	
Asteromonas Artari	cone	5
Aulacomonas Skuja	prolate spheroid	
Botryococcus Kütz.	ellipsoid	
Brachiomonas Bohl.	2 different cones	
Carteria Diesing	sphere	1,5
Characium Braun	ellipsoid	
Chlamydocapsa Fott	prolate spheroid	
Chlamydomonas Ehr.	sphere	5
Chlorangiella De Toni	ellipsoid	

MODELS FOR BIOVOLUME CALCULATIONS OF MICROALGAL TAXA (cont.)

TABLE 2. Continued.

Genus	Shape	A
Chlorangiopsis Korschikoff	ellipsoid	
Chlorella Beijerinck	sphere	
Chlorobion Kors.	cylinder+2 cones	50
Chlorococcum Men.	sphere	5
Chlorogonium Ehr.	2 cones	
Chloromonas Gobi	sphere	5
Chlorotetraëdon McEntee et al.	sphere or box	
Chodatella Lemm.	ellipsoid	
Choricystis (Skuja) Fott	ellipsoid	
Closteriopsis Lemm.	cylinder+2 cones	50
Cocconeis Stein	ellipsoid	
Coelastrum Naeg.	sphere	5
Coenochloris Kors.	sphere	
Coronastrum Thom.	ellipsoid	
Crucigenia Morren	cube	
Crucigeniella Lemm.	cube	
Cystomonas Ettl & Gärtner	ellipsoid	
Dactylosphaerium Steinecke	sphere	1,5
Dermatractum W & GS West	2 cones	
Dichotomococcus Korsikoff	prolate spheroid	
Dictyococcus Gern.	sphere	
Dictyosphaerium Naeg.	sphere	5
Diplochloris Kors.	2 cones	49
Diplostauron Kors.	box	
Dunaliella Teodor.	prolate spheroid	
Emergosphaera Miller	cone+half sphere	
Erballocystis Bohlin	prolate spheroid	
Eremosphaera De Bary	prolate spheroid	
Eudorina Ehr.	sphere	
Fortiella Pascher	ellipsoid	
Franceia Lemm.	prolate spheroid	
Gloeococcus Braun	sphere	
Gloeocystis Naeg.	prolate spheroid	
Gloeomonas Klebs	sphere	5
Golenkinia Chodat	sphere	
Golenkiniopsis Kors.	sphere	
Gonium OF Müller	prolate spheroid	
Granulocystis Hind.	prolate spheroid	
Haematococcus CA Ag.	sphere	
Hafniomonas Ettl & Moestrup	prolate spheroid	
Hormotila Borzi	sphere	
Hyalogonium Pasch.	2 cones	
Hydrianum Raben.	ellipsoid	
Hydrodictyon Roth	cylinder	
Kentrosphaera Borzi	ellipsoid	
Keratococcus Pascher	2 cones	
Kirchneriella Schmidle	2 cones	49
Korshikoviella Silva	2 cones	
Lagerheimia Chodat	ellipsoid	
Lobomonas Dang.	sphere	
Micracantha Kors.	ellipsoid	
Micractinium Fres.	sphere	7
Monoraphidium Komark.-Legn.	2 cones	49
Muriella Boye-Pet.	sphere	
Neochloris Starr	sphere	
Nephrochlamys Kors.	2 cones	49
Oltmannsiella Zim.	prolate spheroid	
Oltmannsiellopsis Chihara & Inouye	prolate spheroid	
Oocystis Braun	prolate spheroid	
Palmella Lyngbye	sphere	
Palmellopsis Kors.	sphere	
Palmodictyon Kütz.	sphere	
Pandorina Bory	prolate spheroid	
Paradoxia Svirenko	2 ellipsoids	
Pascherina Silva	prolate spheroid	
Pediastrum Meyen	elliptic prism	51
Phacotus Perty	ellipsoid	
Planktosphaeria GM Smith	sphere	
Planophila Gerneck	cone+half sphere	
Pleodorina Shaw	sphere	
Podohedra Düringer	2 cones	49
Provasoliella Loebl.	prolate spheroid	
Pseudocarteria Ettl	prolate spheroid	
Pseudocharatium Korsikoff	ellipsoid	
Pteromonas Seligo	ellipsoid	
Pyramichlamys Ettl	ellipsoid	52
Pyrobotrys Arnoldi	prolate spheroid	
Quadrichloris Fott	prolate spheroid	
Quadricoccus Fott	prolate spheroid	
Quadrigula Printz	cylinder + cone	
Raciborskiella Wisl.	cone+half sphere	
Radiosphaera Snow	sphere	
Raphidocelis Hind.	2 cones	49
Scenedesmus Meyen	prolate spheroid	
Scherffelia Pascher	prolate spheroid	
Schroederia Lemm.	2 cones	49
Schroederiella Wol.	ellipsoid	
Scotiella Fritsch	prolate spheroid	
Scotiellopsis Vinatz.	prolate spheroid	
Selenochloris Pasch.	2 cones	
Siderocelis (Naumann) Fott	prolate spheroid	
Sorastrum Kütz.	cylinder + cone	
Sphaerellopsis Korschikoff	prolate spheroid	
Sphaerocystis Chod.	sphere	
Spongiococcum Deason	sphere	5
Tetrachlorella Kors.	prolate spheroid	
Tetracystis Brown & Bold	prolate spheroid	
Tetradesmus GM Smith	2 cones	49
Tetraëdon Kütz.	box	53
Tetraspora Link	sphere	
Tetrastum Chodat	sphere	
Tetratoma Bütschli	prolate spheroid	
Thorakomonas Korschikoff	ellipsoid	
Trebouxia Paymaly	sphere	
Treubaria Bernard	3 cones	
Volvox (L.) Ehr.	sphere	
10c. Zygnematophyceae		
Ancylonema Bergg.	cylinder	
Arthrodesmus Ehr.	2 ellipsoids	
Closterium Nitzsch	2 cones	49
Cosmarium Corda	2 half ellipsoids	
Cosmiocladium De Breb.	2 half ellipsoids	
Cylindrocystis (Menegh.) De Bary	cylinder	
Desmidium CA Ag.	cylinder	
Docidium De Breb.	cylinder	
Euastrum Ehr.	2 truncated cones	
Gonatozygon De Bary	cylinder	
Gymnozyga Ehr.	2 truncated cones	
Hyalotheca Ehr.	cylinder	
Mesotaenium Naeg.	cylinder	
Micrasterias CA Ag.	2 half ellipsoids	54
Mougeoutia CA Ag.	cylinder	
Netrium Naeg.	prolate spheroid	
Onchyonema Wall.	2 ellipsoids	
Oocardium Naeg.	ellipsoid	
Penium De Breb.	cylinder	
Pleurotaenium Naeg.	cylinder	
Sirogonum Kütz.	box	
Sphaerozosma Chorda	2 truncated cones	
Spirogyra Link	cylinder	
Spirotaenia De Breb.	prolate spheroid	55
Spondylosium De Breb.	ellipsoid	
Staurastrum Meyen	2 truncated cones	56
Tetmemoras Ralfs	ellipsoid	
Xanthidium Ehr.	2 half ellipsoids	
Zygnema CA Ag.	cylinder	

MODELS FOR BIOVOLUME CALCULATIONS OF MICROALGAL TAXA (cont.)

TABLE 2. Continued.

1) Cross section may be elliptic rather than round. In this case the squared diameter of the equation should be replaced by the product of smaller x greater diameter.
2) Some cells or species are elongated and should be calculated as cylinders or prolate spheroids.
3) Note that the diameter of the cells changes within one chain.
4) Exceptions: *D. balticum*, *D. cylindricum*: cylinders.
5) Some species are apically elongated. They should be calculated as prolate spheroids.
6) In species which are genuflexed in girdle view, the apical axis can be calculated more precisely, if the length of the two straight parts is summed.
7) Species with a rhombic valve view should be calculated as prisms on a parallelogram.
8) Species with a linear valve view should be calculated as boxes.
9) Some species have great capitate poles, these can be added as cylinders. In this case, the apical axis means the apical length without the capitae.
10) Some species are not in a shape of a lemon wedge, but more like half-elliptic cylinders due to a deep girdle and should be calculated like these. These are e.g. *Amphora decussata*, *A. commutata* or *A. ostrearia*.
11) Height (pervalvar axis) is measured as the mean of the minimum and the maximum.
12) Elevations or extensions (very robust setae, conical apical elevations) should be added separately as cylinders or cones.
13) The smaller part with the head pole is calculated as a square, and the inflated base pole as anelliptic cylinder
14) Some species are round in valve view and can be calculated as cylinders.
15) *Coscindiscus granii* resembles an asymmetric cylinder with added spherical segments and can be calculated as follows:

$$\pi \cdot d^2 \cdot \left(\tfrac{1}{8} \cdot (h + H) + \tfrac{1}{4} \cdot c^3\right)$$

(d= diameter, h= min. height, H= max. height, c= height of sphere segment)

16) Spindle-shaped central part and the rostrate ends are calculated separately.
17) Width (transapical axis) is measured as the mean of the minimum and the maximum.
18) Some species are very weakly heteropolar and can be calculated as elliptic cylinders. These are e.g. *Cymbella lacustris*, *C. aequalis*, *C. cesatii*, *C. amphioxys* or *C. naviculaceae* as well as *Gomphonema insigne* or forms of *G. parvulum*.
19) Species with straight ventral sides should be calculated as half-elliptic cylinders.
20) For some species with inflated central parts, see elliptic prism with transapical inflations.
21) Some species gomphonemoid, see *Gomphonema*.
22) Some forms are more flattened and elliptic prism may fit better.
23) The triangle should be superimposed over half the extension's width.
24) *Melosira nummuloides* is very variable in shape. Often it resembles more a cylinder with 2 half spheres or a sphere.
25) The genus *Nitzschia* is quite variable in its shape. The sigmoid and rhombic cells can be calculated as prism on a parallelogram as described. Elliptic species are to be calculated as elliptic prisms, linear species as boxes. Species of the subgenus *Nitzschiella* should be calculated as proposed in 32.
26) Species with an elliptic valve view should be calculated as elliptic prism.
27) Polymorphic genus with a triangulate form, to be calculated as a prism on triangle.
28) The diameter refers to the inner diameter without corona.
29) If the girdle is very shallow, the volume can be calculated as a sphere.
30) Species with flat valves can be calculated as cylinders.
31) Species with triangulate valves should be calculated as cylinders on triangle.
32) The central inflated part is to be calculated as elliptic prisms, and the two extensions as squares.
33) Some species resemble cones.
34) The euglenoid algae are variable in shape and cross-section (Rott 1981). Most *Euglena sp.* are not round, but flattened in cross-section. Therefore the obtuse pole is calculated as a half ellipsoid, the acute pole as cone with an elliptical base. (Sicko-Goad et al. (1977) propose a similar shape with a cylinder instead of a cone). The smaller and wider diameter have to be measured as well as the height of the cone and the length of the obtuse pole. Some *Euglena sp.* are so flat that they resemble a flat elliptic prism. The genus *Phacus* is leaf-flat (Leedale 1967), sometimes the cells are circular and can be calculated as cylinders. Note, that these elliptic prisms are based on the apical section.
35) Exceptions: *A. concavum*: 2 cones.
36) Exceptions: *A. sphenoides*: prolate spheroid.
37) The genus *Ceratium* is quite variable in shape. The general proposal is: to calculate the central cell body as ellipsoid, then add the hypothecal horns as cones and the apical horn as cylinder. The following exceptions should be considered: a) *C. arcticum*, *C. tripos*, *C. declinatum*, *C. arietinum*, *C. contortum*, *C. concilians*, *C. incisum*: the central body calculated as cone; b) *C. carolinianus*, *C. cornutum*: the apical horn calculated as cone; c) *C. vultur*, *C. trichoceros*, *C. macroceros*, *C. carriense*: all horns calculated as cylinders; d) *C. gravidum*, *C. praelongum*: ellipsoid with 2 cones; e) *C. fusus*, *C. inflatum*: 2 cones.
38) Exceptions: *D. pulchella*: prolate spheroid; *D. tripos*, *D. caudata*: horns have to be added as cones.
39) Exceptions: *G. grindleyi*, *G. fragilis*, *G. alaskensis*: ellipsoid. *G. spinifera*, *G. scrippsae*: cone + half sphere.
40) Exceptions: *G. acuminatum*, *G. rhomboides*: 2 cones. *G. lantzschii*, *G. triceratium*: cone + half sphere.

MODELS FOR BIOVOLUME CALCULATIONS OF MICROALGAL TAXA (cont.)

TABLE 2. Continued.

41) Exceptions: *G. lachrymae*: cone + half sphere.
42) Exceptions: *K. asymmetricum*: ellipsoid. *K. mazaricum*: cylinder + half sphere.
43) Exceptions: *O. splendidens*, *O. heteroporus*: prolate spheroid.
44) Exceptions: *P. achromaticum*: 2 cones. *P. limbatum*: add apices as cones.
45) Exceptions: *P. arcuatum*, *P. micans*, *P. dentatum*, *P. triestinum*: cone + half sphere.
46) Exceptions: a) *P. thorianum*, *P. pellucidum*, *P. punctulatum*, *P. leonis*, *P. brevipes*, *P. obtusum*, *P. pallidum*: cone + half ellipsoid; b) *P. oceanicum*, *P. oblongum*, *P. divergens*, *P. fatulipes*: as a), but with prominent hypothecal horns, which have to be added as cones; c) *P. subinerme*, *P. ovatum*, *P. quarnerense*: ellipsoid; d) *P. elegans*: cylinder + 3 cones; e) *P. minutum*, *P. nudum*: sphere.
47) Cell body as cone, "legs" as cylinders.
48) Exceptions: *A. aciculare*: cone + half sphere, *A. fluviatile*: prolate spheroid.
49) These genera include some species which are straight and others which are bent. The latter cells should be calculated as Monoraphidioids.
50) Some species are not linear but lanceolate in apical section and should be calculated as 2 cones, e.g. *Chlorobion lunulatum* or *Chlosteriopsis longissima*.
51) Elliptic prism refers to the colony of *Pediastrum*, not to single cells.
52) Exceptions: *P. conica*, *P. salina*: cones.
53) Exceptions: *T. trigonum*: pyramid.
54) Some species of *Micrasterias* are deeply divided and can only be estimated by separating the cell into cylinders.
55) Exceptions: *S. condensata*: cylinder.
56) Exceptions: *S. teliferum*, *S. aculeatum*, *S. punctulatum*, *S. dickiei*: 2 ellipsoids.

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