AOS PROTOCOL AND PROCEDURE: ZOOPLANKTON SAMPLING IN LAKES

PREPARED BY	ORGANIZATION	DATE
Charlotte Roehm	AQU	11/14/2012
Stephanie Parker	AQU	08/04/2014

APPROVALS	ORGANIZATION	APPROVAL DATE
Mike Stewart	PSE	08/26/2014

RELEASED BY	ORGANIZATION	RELEASE DATE
Stephen Craft	PSE	08/26/2014

See configuration management system for approval history.

© 2014 NEON Inc. All rights reserved.

The National Ecological Observatory Network is a project solely funded by the National Science Foundation and managed under cooperative agreement by NEON, Inc. Any opinions, findings, and conclusions or recommendations expressed in this material are those of the author(s) and do not necessarily reflect the views of the National Science Foundation.

Change Record

REVISION	DATE	ECO #	DESCRIPTION OF CHANGE
A	05/28/2014	ECO-01127	Initial release
B	08/26/2014	ECO-02238	Minor updates based on feedback from the field

TABLE OF CONTENTS

LIST OF TABLES AND FIGURES

1 DESCRIPTION

1.1 Purpose

The primary purpose of this document is to provide a change-controlled version of Observatory protocols and procedures. This document provides the content for training and field-based materials for NEON staff and contractors. Documentation of content changes (i.e. changes in particular tasks or safety practices) will occur via this change-controlled document, not through field manuals or training materials.

This document is a detailed description of the field data collection, relevant pre- and post-field tasks, and safety issues as they relate to this procedure and protocol.

1.2 Scope

This document relates the tasks for a specific field sampling or laboratory processing activity and directly associated activities and safety practices. This document does not describe:

- General safety practices
- Site-specific safety practices
- General equipment maintenance

It does identify procedure-specific safety hazards and associated safety requirements.

1.3 Acknowledgements

This protocol is based on modified version of U.S. Environmental Protection Agency (2012a, b), Baker et al. (1997), and U.S. Environmental Protection Agency (2009).

2 RELATED DOCUMENTS AND ACRONYMS

2.1 Applicable Documents

Applicable documents contain information that shall be applied in the current document. Examples are higher level requirements documents, standards, rules and regulations.

AD [01]	NEON.DOC.004300	EHS Safety Policy and Program Manual
AD [02]	NEON.DOC.004316	Operations Field Safety and Security Plan
AD [03]	NEON.DOC.000724	Domain Chemical Hygiene Plan and Biosafety Manual
AD [04]	NEON.DOC.001155	NEON Training Plan
AD [05]	NEON.DOC.050005	Field Operations Job Instruction Training Plan
AD [06]	NEON.DOC.001197	Bathymetric Sampling of Lakes and Non-Wadeable Streams

© 2014 NEON Inc. All rights reserved.

2.2 Reference Documents

Reference documents contain information complementing, explaining, detailing, or otherwise supporting the information included in the current document.

RD[01]	NEON.DOC.000008	NEON Acronym List
RD[02]	NEON.DOC.004300	EHS Safety Policy and Program Manual
RD[03]	NEON.DOC.005003	NEON Scientific Data Products Catalog
RD[04]	NEON.DOC.014051	Field Audit Plan
RD[05]	NEON.DOC.001152	NEON Aquatic Sample Strategy Document
RD[06]	NEON.DOC.000824	Data and Data Product Quality Assurance and Control Plan
RD [07]	NEON.DOC.001154	NEON Aquatic Decontamination Protocol
RD [08]	NEON.DOC.001646	NEON General AQU Field Metadata Sheet
RD [09]	NEON.DOC.002302	Datasheets for AOS Protocol and Procedure: Zooplankton Sampling in Lakes
RD [10]	NEON.DOC.002191	Datasheets for Secchi Depth and Depth Profile Sampling
RD [11]	NEON.DOC.001164	NEON Bathymetric Mapping Protocol

2.3 External References

External references contain information pertinent to this document, but are not NEON configuration-controlled. Examples include manuals, brochures, technical notes, and external websites.

ER [01]	
ER [02]	
ER [03]	

2.4 Acronyms

DI	Deionized
EMAP	Environmental Monitoring and Assessment Program (USEPA)
mL	milliliter
NLA	National Lakes Assessment (USEPA)
PFD	Personal Flotation Device
USEPA	United States Environmental Protection Agency
USGS	United States Geological Survey

© 2014 NEON Inc. All rights reserved.

2.5 Definitions

A protocol is a formal summary description of a procedure and its related rationale, and includes information on knowledge and resources needed to implement the procedure. A procedure is a set of prescribed actions that must take place to achieve a certain result, and can also be called a method. It differs from a science design in that science designs provide a more complete description of the rationale for selecting specific protocols. It differs from a training manual in that training manuals provide materials in support of skills acquisition in the topic areas including information on how to best train staff rather than detailing only the steps of the procedure.

Euphotic zone: The upper layer of lake water where sunlight penetrates and photosynthesis can occur. Usually demarcated by the 1% light penetration level.

Eutrophic : The ecosystem having high primary production. In lakes, this often leads to increased production of algae or algal blooms.

Epilimnion: The upper layer of the lake that overlies the thermocline. This layer tends to be less dense with a higher pH and oxygen concentrations.

Hypolimnion: The dense bottom layer of a stratified lake that sits below the thermocline. This layer is denoted by cooler summer temperatures and slightly warmer winter temperatures relative to the Epilimnion.

Metalimnion: The layer of water in a stratified lake that sits between the hyplolimnion and the epilimnion. Often equated with the thermocline (Figure 2).

Oligotrophic: The ecosystem response to low nutrient content. In lakes, this often equates to very clear water and little algal production.

Pelagic: The part of the lake that is not near shore or close to the bottom.

Stratified: Indicating the presence of a thermocline.

Thermocline: A distinct layer in a body of water where the change in temperature is more rapid than increasing depth. The denser and cooler layer below the thermocline is defined by the hypolimnion. The warmer upper layer is termed the epilimnion.

© 2014 NEON Inc. All rights reserved.

3 BACKGROUND AND OBJECTIVES

3.1 Background

Zooplankton inhabit all layers of a water body and constitute a major link between primary production and higher trophic levels in aquatic ecosystems. Unlike algae or phytoplankton, zooplankton are microscopic animals that do not produce their own food (Fig. 1). These small invertebrates float freely in the water column of lakes and oceans. They are important as both prey and consumers in the aquatic food web. Zooplankton are filter feeders that feed primarily on algae whilst also being the primary source of food for planktivorous fish. Zooplankton assemblage responds to environmental stressors such as nutrients, and such effects can be detected through changes in species composition, abundance, and body size distribution (U.S. Environmental Protection Agency, 2012).

Many zooplankton are capable of strong swimming movements and may migrate vertically from tens to hundreds of meters; others have limited mobility and depend more on water turbulence to stay afloat. Zooplankton can be classified according both to developmental stages: Meroplankton and holoplankton; and to size (Sanders and Porter, 1990). Meroplankton spend only part of their life cycles as plankton, whilst holoplankton spend essentially their whole existence in the water column. Freshwater zooplankton are dominated by six major groups of animals: protozoa, rotifers, two subclasses of the Crustacea, the cladocerans and copepods and *Mysis* and *Chaoborus* (Fig. 1a-d). The planktonic protozoa in particular, have limited locomotion and are dominated by the meroplanktonic pelagial zooplankton (5 – 300 µm), dominated by a summer planktonic lifecycle, spending the rest of their life cycle in the sediments. Several non-planktonic rotifers (150 µm – 1 mm), are sessile (higher mobility) and mostly associated with the littoral zone (Fig. 2). Most rotifers are nonpredatory, and omnivorously feed on bacteria, small algae, and detrital particulate organic matter. The majority of cladoceran are small (0.2 to 3.0 mm) and have a distinct head and bivalve carapace covering the body. Locomotion is accomplished mainly by means of the large secondary antennae. Planktonic copepods (2-4 mm) consist of two major groups, the calanoids (*Diaptomus*) and the cyclopoids, distinguished by their body structure and length of antennae.

© 2014 NEON Inc. All rights reserved.

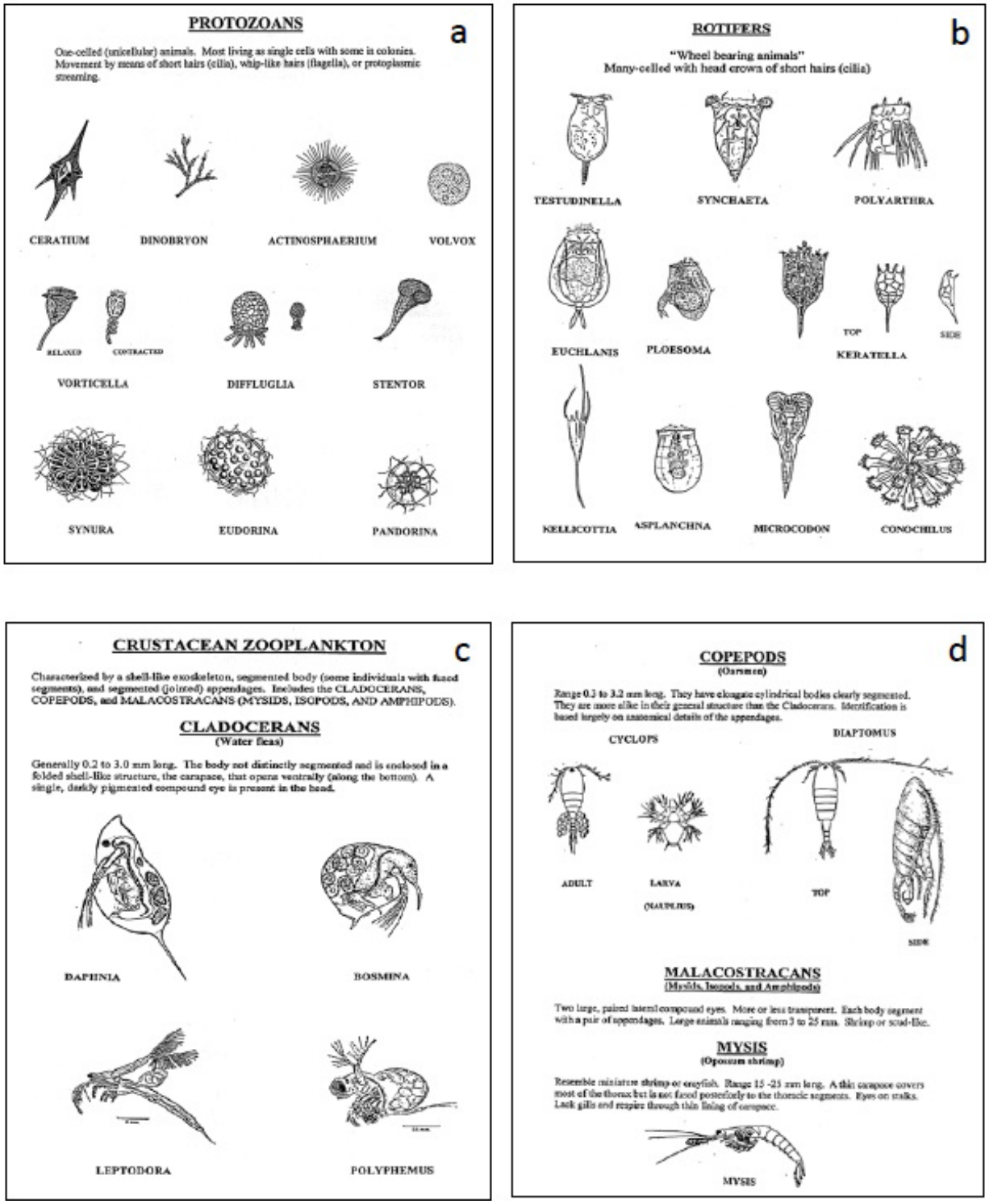

Figure 1. Classes of zooplankton (from Tharp).

© 2014 NEON Inc. All rights reserved.

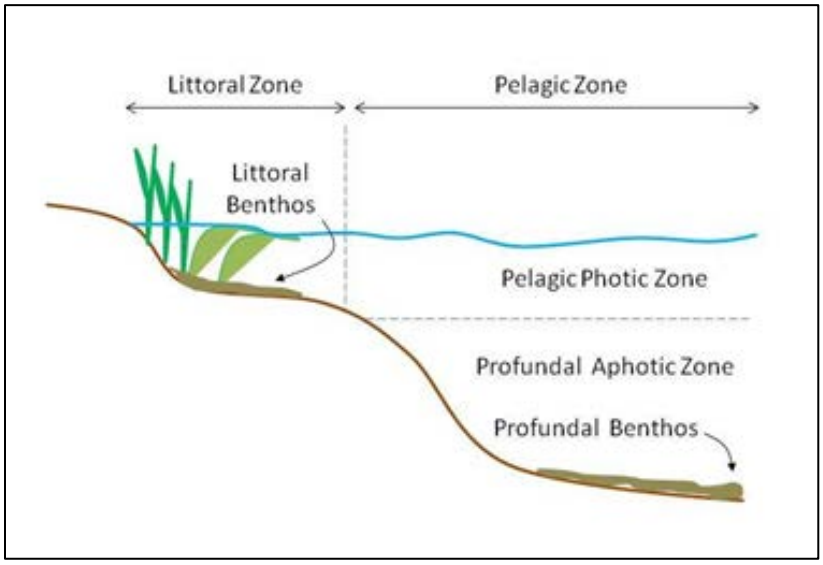

Figure 2. Zones of a lake.

Seasonality plays an important role in zooplankton species presence and abundance, with cyclopoid and calanoid copepods dominate the community in the winter months, with large cladocera peaking in mid-summer and small cladocera in later summer early autumn (Gerten and Adrian, 2002). A change in lake trophic conditions from oligotrophic to eutrophic has been shown to result in size-selective predation that potentially contributes to the decline of larger zooplankton, with ciliated protozoans and rotifers becoming more important (Wetzel, 2001).

3.2 NEON Science Requirements

This protocol fulfills Observatory science requirements that reside in NEON's Dynamic Object-Oriented Requirements System (DOORS). Copies of approved science requirements have been exported from DOORS and are available in NEON's document repository, or upon request.

3.3 NEON Data Products

Execution of this protocol procures samples and/or generates raw data satisfying NEON Observatory scientific requirements. These data and samples are used to create NEON data products, which are documented in the NEON Scientific Data Products Catalog, available on the NEON website.

© 2014 NEON Inc. All rights reserved.

4 PROTOCOL

The goals of the Zooplankton Sampling in Lakes Protocol are to quantify biodiversity, number of species present, and biomass (or biovolume) at each lake site. These variables will be used to build a database over time, in order to track changes in zooplankton community structure and function, and introduction of invasive species.

This protocol is based on modified version of U.S. Environmental Protection Agency (2012), Baker et al. (1997), and U.S. Environmental Protection Agency (2009).

Samples shall be collected as an integrated water column sample at the central location of the lake. Two additional samples are taken at the inlet and outlet of the lake or a location downwind of the predominant wind direction. Samples are taken 3 times per year in order to capture multiple species presence and abundance.

5 QUALITY ASSURANCE AND CONTROL

The procedures associated with this protocol will be audited according to the Field Audit Plan (RD[04]). Additional quality assurance will be performed on data collected via these procedures according to the NEON Data and Data Product Quality Assurance and Control Plan (RD[06]).

If conditions become too unsafe to continue working before all samples have been collected, discard collected samples and return to the shore. Rinse all equipment thoroughly. Return to sample after conditions have stabilized.

When unexpected field conditions require deviations from this protocol, the following field implementation guidance must be followed to ensure quality standards are met:

1) All samples from one sampling bout must be collected within one day (i.e., all samples per lak/non-wadeable stream as detailed in this protocol).
   a) If circumstances occur that impede sampling (e.g., wildlife, weather), start sampling again the next day that conditions permit.
   b) If circumstances occur that delay sampling (e.g., lightning), but sampling can be continued the same day while still meeting the streamflow requirements below, continue to collect samples.
2) Samples shall be taken a minimum of 5 days following a major flow event (>25% change in flow within 15 minutes and/or turbidity levels are double the monthly average), allowing for recolonization before sampling occurs.
3) If weather conditions deteriorate and the lake/non-wadeable stream becomes too windy (>9 km hr^{-1}) to hold the boat stationary over a sampling point, return to shore and wait in a safe location for 30 minutes. If wind subsides, resume sampling, if not, return to the Domain Support Facility and sample at another time.
   a) If you are able to return to the lake/non-wadeable stream to sample within 24 hours, you may keep samples from the previous day. If you are not able to return within 24 hours, discard any previously collected samples in the lake/non-wadeable stream or at the Domain Support Facility and start over.
4) A minimum of 2 weeks between sample periods shall be observed.

© 2014 NEON Inc. All rights reserved.

5.1 Sampling-Specific Concerns

1) Zooplankton often become trapped in the folds of the nets (near the stitching). Check net seams between samples to ensure that specimens are added to the correct samples, and do not remain in the net between sites.
2) Samples must be preserved within 30 minutes in the field.

6 SAFETY

Personnel working at a NEON site must be compliant with safe field work practices as outlined in the Operations Field Safety and Security Plan (AD[02]) and EHS Safety Policy and Program Manual (AD[01]). Additional safety issues associated with this field procedure are outlined below. The Field Operations Manager and the Lead Field Technician have primary authority to stop work activities based on unsafe field conditions; however, all employees have the responsibility and right to stop their work in unsafe conditions.

See Section 10 in the NEON Operations Field Safety and Security Plan (AD 01]) for aquatic-specific field safety requirements. In addition, the following safety requirement must be followed:

1) Due to site-specific hazards that may be encountered, sampling may have to take place from the boat, without dismounting from the vessel. In addition, technicians are required not to put hands and feet in waters where alligators are present and to make sure a safe distance from hazards is maintained.
2) All personnel must be wearing a personal flotation device (PFD) prior to entering the boat.
3) All employees shall have access to a form of communication with other team members such as a two-way radio.
4) Technicians should be aware of any site-specific hazards and to the waters of that particular location (i.e. current status, tidal charts, etc.)

7 PERSONNEL REQUIREMENTS

Field sampling requires two technicians for four hours each sampling day plus travel to and from the site.

Where applicable, personnel will be licensed to operate a boat and able to safely handle a motor and drive a boat safely.

8 TRAINING REQUIREMENTS

All technicians must complete required safety training as defined in the NEON Training Plan (AD[04]) and EHS Safety Policy and Program Manual(AD[01]). Additionally technicians complete protocol specific training for safety and implementation of protocol as required in Field Operations Job Instruction Training Plan (AD[05]). Personnel are to be trained in the field protocols associated with this document, and trained in safe working practices for lake-based fieldwork.

© 2014 NEON Inc. All rights reserved.

9 FIELD STANDARD OPERATING PROCEDURE

9.1 Sampling Frequency and Timing

Ranges of sample timing are provided on a site-by-site basis based on metadata collected by the aquatic sesnors and field technicians. See the NEON Aquatic Sample Strategy Document (RD [05]) for ranges of sampling dates.

9.1.1 Criteria for Determining Sampling Dates

A range of dates for each site will be determined *a priori*, based on historical data including ice on/ice off, the accumulation of degree days, weather, and riparian phenology

9.1.2 Sampling Frequency

Lake zooplankton sampling occurs three times per year at each site, roughly spring, summer, and autumn.

9.1.3 Sampling Timing Parameters

Sampling must occur within a one month window of the determined sampling date, depending on weather conditions at the site, with a minimum of two weeks between sampling dates.

© 2014 NEON Inc. All rights reserved.

9.2 Equipment and Materials

Table 1. Field equipment list

Maximo Item No.	Item Description	Quantity	Habitat-Specific	Special Handling
General equipment				
	Site-specific bathymetry map (RD [11])	1		
	Aquatic Field Metadata Sheet, RD [08] (all-weather paper)	1		
	Field data sheets (all-weather paper, RD [10])	2		
	Pre-printed adhesive labels	1 sheet		
	Laboratory nitrile gloves	3 pair		
	Work gloves	1 pair		
	Pencils	4		
	Permanent markers	4		
	Metric ruler	1		
	Calculator	1		
	Tape (roll)	1		
	Cooler with ice packs	1		
Secchi depth and Depth Profile equipment				
MX100453	GPS and Sounder Unit (Humminbird)	1		
	Sounder Battery	1		Y
MX100450	Secchi disk and attached rope	1		
MX100512	Multisonde for temperature and depth	1		
Sampling equipment				
MX100397	Plankton tow net 63 μm pore size with collection bottle	1		
MX105580	Schindler-Patalas sampler 64 µm pore size	1		
	Nylon safety line for net (10-50 m)	1		
	500 mL Nalgene sample bottles	6		
	500 mL Wash bottle	1		
	70% Ethanol	1		Y
	50-100 mL graduated cylinder	1		
	Plastic sampling tray	1		
	Forceps	1		
	Resealable bags (gallon)	3		
General boating equipment				
	Boat, Oars and Electric Motor	1		
	Anchor and line	1		
	Personal flotation devices (PFDs), 1 per person	2		
	Safety kit for boat	1		
	First aid kit	1		

© 2014 NEON Inc. All rights reserved.

9.3 Preparation

1) Collect and prepare all equipment, including sample bottles and labels.
   a) Load GPS sampling coordinates in GPS.
2) Pre-print adhesive labels for sample bottles and data sheets on all-weather paper.
3) Check for holes in nets, assure that nets are clean and free of debris and organic matter and have been decontaminated (RD[12]) since last use.
4) Have ice or ice packs frozen and ready for cooler.
5) Check that all equipment is in good condition and all batteries are charged.

9.4 Sample Collection in the Field

1) Locations for sampling:
   a) In the deepest point in the lake, determined by site map and pre-loaded GPS coordinates, 2 integrated samples.
      a) Location ID = “center”
   b) Near the major inlet in the littoral zone, 2 integrated samples.
      a) Location ID = “inlet”
   c) Near the outlet in the littoral zone, 2 integrated samples.
      a) Location ID = “oulet”
2) Ensure the General AQU Field Metadata Sheet (RD [08]) is completed.
3) Navigate the boat to the central sampling location as defined by bathymetric maps of the lake [RD [11]]
4) Fill in the location information on the field sheet (RD [09]) and note the GPS position of the sampling location.
5) Gently lower the anchor to the bottom of the lake so as not to suspend sediments.
6) Always sample near the bow of the boat to minimize the effects of the motor on the water column. When anchored, the bow of the boat tends to orient itself with the bow into the wind or current.
7) Determine the total water depth from the sonar readings.
8) Determine the depth of the thermocline (if present) at the sampling location using the multisonde. Record the depth and water temperature every 0.5 m on the Depth profile field data sheet (RD [10]).

© 2014 NEON Inc. All rights reserved.

NEON Zooplankton Collection

Lakes

Site (4-letter code):	**Recorded by:**
Date (YYYYMMDD):	**Collected by:**
Local time (HH:MM):	**Sampling protocol & Rev.:**

Sampling locations

Location ID	Latitude	Longitude

Location ID	Sample type	Replicate	Sample ID	Sample depth (m)	Number of tows

Figure 3. Example field sheet for zooplankton collection. (Blank sheet in RD [11]).

9) Determine the depth of the euphotic zone (depth to which 1% light penetrates) using the Secchi disk (Figure 4, RD [10]).
   a) Take the Secchi reading from the shady side of the boat. If there is no shade use a hand or any object that can produce enough shade to cut down the sun glare. DO NOT WEAR SUNGLASSES WHEN UNDERTAKING THIS MEASUREMENT.
   b) Lower the Secchi disk slowly until the white quadrants disappear, and determine the depth to the nearest tenth of a meter.
   c) Lower the Secchi disk so it is about 0.5 m below the first reading. Slowly pull the disk up until it reappears, and determine this depth to the nearest tenth of a meter.
   d) Average the two depths to get the Secchi depth. Record all readings on the data sheet.
   e) Repeat at each sampling location.

© 2014 NEON Inc. All rights reserved.

Figure 4. Taking a Secchi Disk measurement on the shaded side of the boat.

10) Determine which zooplankton sampler to use.
    a) If water depth is < 2 meters use a Schindler-Patalas sampler.
        1) If <2 but > 1 m take samples at 0.5 m and 1.5 m depths and integrate.
        2) < 1 m take 2 samples from 0.5 m.
    b) If water is > 2 meters use a plankton tow net sampler to take an integrated sample.
    b) Note the sample type field data sheet (RD [11]).
11) If using a tow net (Figure 5).
    a) Attach the net to a nylon rope marked at 0.5 m intervals.
    b) Lower the net to where the bottom tubing is 0.5 m above the bottom of the lake. If conditions are windy or conditions necessitate, add a small weight at the bottom of the net to help lower the netting.
    c) Pull the nylon rope vertically at a rate of 0.5 m/s (net tow).
    d) Inspect the sample for organic and inorganic debris (leaves, sediments etc.). If excessive debris is noted, discard sample and start again.
    e) If algae clogs the net, rinse the sides of the net down with the 500 mL wash bottle. If water cannot escape from the net, consider using the Schindler-Patalas sampler at this location.
    f) After the inspection, slowly lower the net into the lake to within inches of the net opening above the water surface and abruptly pull upwards out of the water to collect the contents within the net into the collection cup. *Take care not to submerse the top of the net opening below the water surface*. Repeat several times until the net is fully rinsed.

© 2014 NEON Inc. All rights reserved.

Figure 5. Zooplankton Tow Net

g) Complete the rinsing of the net contents by spraying water against the outside of the net with a wash bottle filled with lake water.

h) Holding the net in a vertical position, carefully open the spigot on the hose and pour into the 500 mL sample bottle.
   a) If debris is present either discard the whole sample (i.e. lots of particulate debris) or pour the sample into the plastic tray and remove the large debris with forceps.

i) Rinse the concentrated zooplankton into the collection bottle using a squirt bottle filled with lake water.

j) The total sample in the bottle should be 1/3 full with zooplankton and water.

k) Note the tow depth (distance in meters of water column through which the collecting unit was retrieved) on the field data sheet(Figure 6, RD [11]) so sample volume can be calculated.

l) Rinse the net, bottle and retrieval rope thoroughly (2 to 3 times) with water from the site.

m) Repeat tows (Steps b-n) until the sample bottle is ¾ full (allow enough space for 40 mL of preservative).

n) In shallow lakes, multiple tows with each net are required to achieve the cumulative tow length representative of the water column.

o) Note the number of tows on the data sheet (RD [11]) in order to calculate total sample volume)

p) Preserve according to Sample Preservation (Section 9.5) and place sample bottle in cooler.

q) Proceed to the next sample location, and rinse the net, collection cup and retrieval rope thoroughly (2 to 3 times) with water from the site on the downwind side of the sampling location and repeat above steps.

© 2014 NEON Inc. All rights reserved.

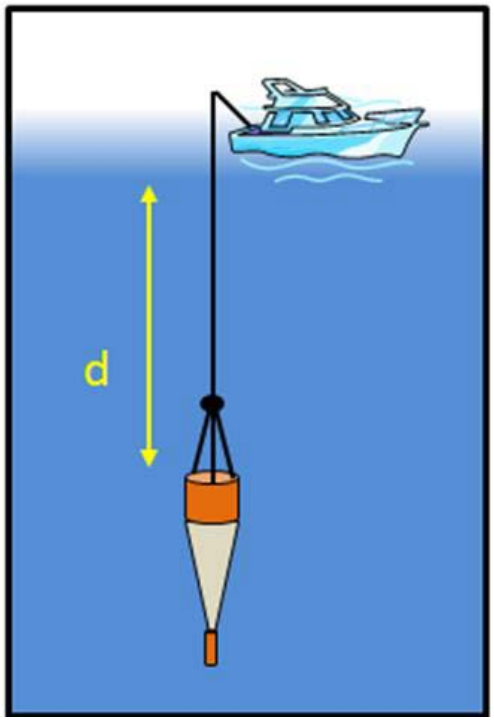

Figure 6. Tow volume calculation

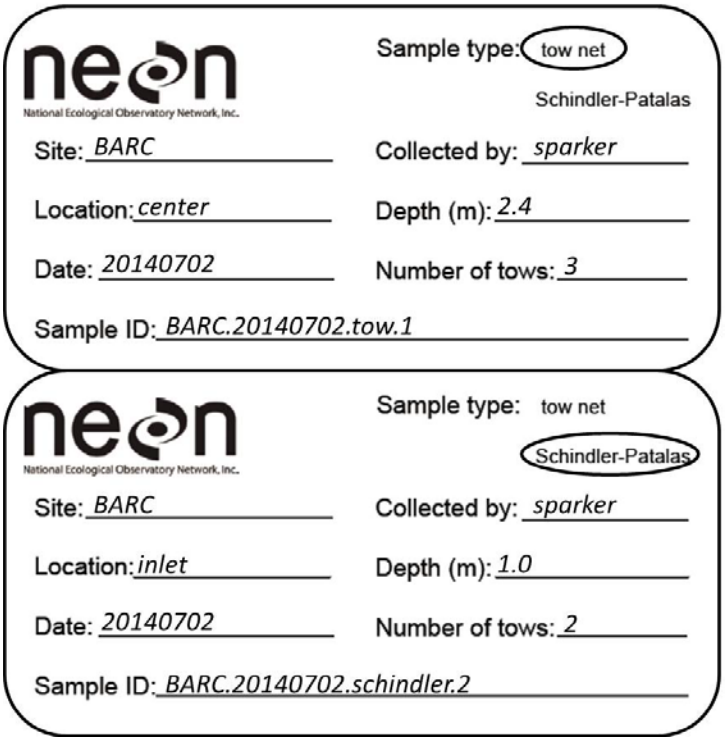

Figure 7. Example of field labels for zooplankton sample bottles.

© 2014 NEON Inc. All rights reserved.

12) If a Schindler-Patalas sampler is used (Fig. 8):
   a) Ensure the dolphin bucket is attached.
   b) Lower sampler to appropriate depth.
   c) When the depth is reached the trap doors will close automatically.
   d) Bring the sampler to the surface. The water will drain through the net.
      a) When filled with water, this sampler is heavy. Work gloves may be worn to protect hands.
   e) Rinse the sampler and net with the 500 mL wash bottle filled with lake water until all samples is inside the dolphin bucket.
   f) Carefully remove the dolphin bucket and transfer sample to a 500 mL sample bottle. Rinse dolphin bucket into collection bottle.
   g) Repeat steps above to integrate two Schindler-Patalas samplers.
   h) Preserve according to Sample Preservation (Section 9.5) and place sample bottle in cooler.
   i) Proceed to next sampling location and repeat steps above.

Figure 8. Example of a Schindler-Patalas sampler

© 2014 NEON Inc. All rights reserved.

9.5 Sample Preservation

All processing must be completed in the field. Should conditions not allow for this step to be undertaken in the field within 5 minutes of sampling, all preservatives shall be added within 30 minutes of collecting the sample in the field and noted in the comments section.

1) Put on Nitrile gloves
2) Add 40 mL of 70% ethanol (or dilute 95% ETOH) solution to the sample within 5 minutes of collection (final concentration ~30-40% ETOH).
3) Carefully re-cap bottle, ensuring that no sample escapes and gently invert bottle to mix.
4) Store in a cooler to return to Domain Support Facility.

9.6 Sample Shipping

1) Samples may be stored at the domain support facility at 4 ^{o}C until shipping.
    a) Shipping should occur within one week of sampling, however samples may be held for up to 30 days at the domain support facility if necessary.
2) Tape bottle lids and ship samples at ambient temperature.
3) Follow shipping and Haz-mat procedures for ethanol.
    a) Place package inside appropriate sturdy shipping container. Add absorbent packing material as necessary to take up excess space in container.
4) Include return shipping label if any shipping materials that need to be returned to the domain support facility (e.g., cooler).
5) Include sample shipping inventory (see Excel workbook “AOS sample shipping inventory”).
    a) Email shipping inventory to external lab contact and copy the NEON CLA contact.
6) Tape and label container for shipping and ship to zooplankton taxonomy lab

9.7 Data Handling

1) Enter all data from the Lab Data Sheets into Excel workbooks “lake_zooplankton data entry” within 72 hours of processing. Follow NEON manual data handling procedures.
2) Archive/file all paper field sheets at the Domain Support Facility or with NEON Cyber-Infrastructure.
3) Upload data to the NEON Cyber-Infrastructure database.

9.8 Refreshing the Sampling Kit

1) Replace sample bottles.
2) Print new field labels and field data sheets.
3) Refill/restock preservative containers.

© 2014 NEON Inc. All rights reserved.

9.9 Equipment Maintenance, Cleaning, and Storage

1) Decontaminate all equipment that has come in contact with lake/non-wadeable stream water according to the NEON Aquatic Decontamination Protocol (RD 08)].
2) Dry all equipment thoroughly before storage.
3) Check all nets for holes and patch if necessary.

© 2014 NEON Inc. All rights reserved.

10 REFERENCES

Baker, J.R., D.V. Peck, and D.W. Sutton (Eds.) 1997. Environmental Monitoring and Assessment Program Surface Waters: Field Operations Manual for Lakes. EPA/620/R-97/001. U.S. Environmental Protection Agency, Washington, D.C.

Gerten, D. **a**nd Adrian, R. 2002. Species-specific changes in the phenology and peak abundance of freshwater copepods in response to warm summers. *Freshwater Biology* 47, 2163–2173.

Sanders, R.W. and K.G. Porter. 1990. Bacterivorous flagellates as food resources for the freshwater crustacean zooplankter *Daphnia ambigua*. Limnol. Oceanogr. 35: 188-191.

Tharp, R. http://gvsu.edu/wri/education/instructor-s-manual-appendix-d-44.htm.

U.S. Environmental Protection Agency. 2005. Standard Operating Procedure for Zooplankton, Revision 10, (http://www.epa.gov/glnpo/monitoring/sop/chapter_4/LG402.pdf).

U.S. Environmental Protection Agency Minnesota. 2009. National Lake Assessment (NLA) protocol. National Lakes Assessment: a collaborative survey of the Nation's lakes. EPA 841/R-09/001, U.S. Environmental Protection Agency, Office of Water and Office of Research and Development, Washington, D.C.

U.S. Environmental Protection Agency. 2012a. National Lakes Assessment Program, Field Operations Manual.

U.S. Environmental Protection Agency. 2012b. Sampling Procedures for the Great Lakes.

Wetzel R G. Limnology. Philadelphia: Saunders, 2001.

© 2014 NEON Inc. All rights reserved.