AQUATIC PLANT COMMUNITY SAMPLING PROCEDURE FOR DEPRESSIONAL WETLAND MONITORING SITES

I. PURPOSE

To describe and document the standard operating procedure (SOP) used by the Minnesota Pollution Control Agency's (MPCA) Biological Monitoring Program to collect aquatic plant community information at depressional wetland monitoring sites for the purpose of assessing water quality and developing biological assessment criteria.

II. SCOPE/LIMITATIONS

The following SOP applies to all depressional wetland monitoring sites for which an integrated assessment of water quality is to be conducted. An integrated depressional wetland assessment involves the collection of biological (macroinvertebrate and plant) and chemical data to assess wetland condition. The MPCA defines depressional wetlands as wetlands that occur within a shallow depression in the landscape that are not directly associated with streams (i.e., riparian wetland) or lakes (i.e., lacustrine fringe wetland); have a semi-permanent to permanent flooding regime (i.e., not temporarily flooded wetland or vernal pool); and have predominantly emergent marsh to shallow open water (aquatic) vegetation types (Eggers and Reed 1997). This combination of water regime and vegetation communities corresponds to U.S. Fish and Wildlife Service (US FWS) Circular 39 wetland types 3, 4, and 5 (Shaw and Fredine 1956).

III. GENERAL INFORMATION

Sites may be selected for assessment for a number of reasons including: 1) sites randomly selected for ambient condition monitoring, 2) sites selected for the development and calibration of biological criteria, 3) sites selected to evaluate a suspected source or result of pollution impacts, and 4) wetland management/restoration/remediation effectiveness monitoring. Although the reasons for monitoring a site may vary, the aquatic plant sampling protocol described in this document applies to all MPCA wetland monitoring sites unless otherwise noted.

IV. PERSONEL REQUIREMENTS

A. Field Crew Leader: The field crew leader must be a professional aquatic biologist with a good knowledge of the Minnesota wetland flora. He or she must have a minimum of a Bachelors degree in aquatic biology, botany, or a closely related field; and have a minimum of six months field experience in wetland plant sampling and plant identification. Field crew leaders should also be proficient with map reading and orienteering; using both Global Positioning System (GPS) and compass.

B. Field Assistant/Intern: The field assistant/intern must have at least one year of college education and an interest in aquatic biology. Coursework in environmental, natural resource, and/or biological science is preferred.

C. General Qualifications: All personnel conducting this procedure must have the ability to perform rigorous physical activity in an outdoor setting; be capable of lifting up to 50 lbs. of sampling equipment; be able to travel up to four nights per week during the summer months; and maintain a positive attitude within a team setting.

V. RESPONSIBILITIES

A. Field Crew Leader: The field crew leader is responsible for implementing the action steps of the procedure and ensuring that the data generated meets the standards and objectives of the Biological Monitoring Program and the MPCA. In addition, the field crew leader is responsible for planning sampling activities and ensuring that MPCA policies are followed during all sampling activities.

B. Field Assistant/Intern: The field assistant/intern is responsible for implementing the action steps of the procedure; including the maintenance, stocking, and storage of sampling equipment, data collection, and data recording.

VI. TRAINING

All personnel will receive instruction from a trainer designated by the program manager. Major revisions in this protocol require that all personnel that apply this procedure on behalf of the MPCA Biological Monitoring Unit be re-trained in the revised protocol by experienced personnel. The field crew leader will provide additional instruction to the field assistant/intern and will be responsible for monitoring the performance of the field assistant/intern throughout the field season.

VII. ACTION STEPS

A. Equipment Check: Before heading out into the field, check all equipment and supplies necessary to complete this procedure is present and in proper working condition (Table 1).

B. Field Sampling: The wetland vegetation biological assessment techniques employed by the MPCA (i.e., Index of Biological Integrity; Gernes and Helgen 2002, Genet et al. 2006) require data on the different kinds of plants growing in a wetland and how abundant those plants are. The vegetation sampling technique described in this procedure is adapted from what is known as releve sampling. Releve sampling was developed by Braun-Blaunquet in Europe and is currently being used by the Minnesota Department of Natural Resources (DNR) County Biological Survey and Natural Heritage Programs (Almendinger 1987). Essentially, releve sampling relies on the observer to select areas within the desired community that are representative of the overall community composition to place a sampling plot where plant data can be quantified.

Table 1. Equipment List-This table identifies all the equipment needed to complete the MPCA wetland vegetation sampling protocol.

Equipment	Purpose	Operation Check
Personal Data Assistant (PDA)	-Field data recording device	-Software function -Date and time -Associated cords and devices
Global Positioning System (GPS)	-Navigation and sample location recording	-Date and time -Correct coordinate system and datum -Associated cord
Laptop Computer	-Downloading and data storage -GIS applications	-Software function -Associated cords and devices -Power inverter
Digital Camera	-Photographic site documentation	-Memory card(s) -Associated cords -Date and time
Cell Phone	-Communication	-Associated cord
4-8Rechargeable AA Batteries & Charger	-Spare batteries for GPS and digital camera	
Site Files and Maps	-Site location information	
Paper Data Sheets & Clipboard	-Backup in case of PDA failure	
Field Notebook	-Recording misc. notes, backup for recording data	
6 Tall Garden Stakes	-Sampling plot corner posts, 2-spares	
4 50 m Measuring Tapes	-For laying out sampling plots, 2-spares	
Chest Waders	-To keep field workers dry	
Raingear	-To keep field workers dry	
Field Guides	-Aid with plant identification	
Hand Lens	-Aid with plant identification	
1-2 Gallon Size Plastic Bags	-For collecting plant specimens	

Table 1. Equipment List-Continued.

Equipment	Purpose	Operation Check
Cooler with Ice	-Short term preservation of water quality samples (*see Wetland Water Chemistry SOP*) and plant specimens	
Plant Press with Newsprint, Blotters, & Cardboard	-For pressing plant specimens	
Wax Paper	-Aid with pressing aquatic plant specimens	
Shallow Pan	-Aid with pressing aquatic plant specimens	
Compass	-Navigation & sampling plot layout	
Pencils	-For recording data	
Permanent Marker	-For labeling bags & water samples (*see Wetland Water Chemistry SOP*)	
First-Aid Kit	-Emergency medical care	

Since 2001, the MPCA has been collecting field plant data using a hand held Personnel Data Assistant (PDA). This has reduced the amount of time recording data in the field, increased data quality control, and also reduced the time needed to produce sample results. Field data sheets continue to be maintained, however, as a backup to the PDA and are included at the end of this SOP. The MPCA is currently using Trimble Recon™ PDAs with a custom data recording application adapted from the field data sheets and built-in GPS receivers.

B.1. Record visit information: Upon arrival at a site begin recording visit information on the Visit Data Sheet (*attached at the end of this SOP*) or PDA. Record the **Site Name**, **Date**, **Surveyor Name**, and **Arrival Time** immediately. Also, document weather conditions in the **Weather Notes** space.

Throughout the remainder of the visit (i.e., during or following vegetation sampling), record other visit or site level data as appropriate. Document any site photographs in the **Photo Information** section. Record the **Camera Make and Model** used for the visit and the **Photo Number** reported from the camera and any associated **Photo Notes** for each photograph taken. Collect water chemistry measurements and samples (*see Depressional Wetland Water Chemistry SOP*), and record information in the **Water Chemistry** section. Also during the visit, conduct a site stressor verification assessment. Do this by walking around the margin of the wetland, noting any anthropogenic stressors that may be impacting the wetland. Complete the **Habitat Alteration**, **Hydrologic Modification**, and **Sedimentation** checklists in the **Site Stressor Verification** section as you

proceed. Site stressor information is necessary for developing a Human Disturbance Score (HDS; Gernes and Helgen 2002) for the site. A brief site stressor assessment may have been completed during the initial site reconnaissance (*see Wetland Site Reconnaissance SOP*). The purpose of the site stressor verification during vegetation sampling is to assess the wetland more thoroughly and add to any information gathered during the site reconnaissance. Finally, record the **Leave Time** (site departure time) when all of the data have been collected.

B.2. Determine the major plant communities in the wetland: The releve sampling method relies on the observer finding a 'representative' location in the wetland that best characterizes the vegetation of the entire wetland to place the sampling plot(s). The first step in this procedure then is to determine what the major plant communities in the wetland are. This can be done by finding an area where the entire wetland can be viewed or by walking around the margin of the wetland.

B.3. Establish the sample location: After the major plant communities have been identified, determine a location where the sampling plot(s) can be placed that would best capture or represent the vegetation types found in the wetland. Typically, this is at the emergent/aquatic vegetation interface (Figure 1). If the wetland has predominantly emergent vegetation, locate the sample plot(s) in the wettest location of the wetland. If there is not an extensive emergent community present, locate the sampling plot(s) where the emergent community should be.

B.4. Determine the plot size and shape: Over the course of the development of wetland vegetation monitoring at the MPCA, the sampling methods have evolved to better characterize wetland vegetation and increase the performance of the assessment indicators. Because of this evolution, a variety of sampling plot sizes and shapes have been, and continue to be, employed with this procedure. Historically, the MPCA used a single large sampling plot to characterize an entire wetland. The size of the plot was standard (100 m^2), but the shape was either square (10 m x 10 m) or rectangular (5 m x 20 m). The 10 m x 10 m plot was used when a wide and well developed emergent vegetation fringe was present. The 5 m x 20 m plot was employed when only a narrow emergent vegetation fringe was present to better capture the emergent/aquatic vegetation interface. More recently, the MPCA has investigated alternative sample techniques. During the 2003 field season, a methods comparison was undertaken comparing the use of the large single plot versus a set of four small (5 m x 5 m) plots sampling technique (Genet et al. 2005). In this scenario the four small plots survey the same area (100 m^2) and together are considered to be a single wetland vegetation sample. The four small plot technique was found to approximately double IBI precision in the North Central Hardwood Forest Ecoregion (Figure 2) and it was decided that it should be adopted as the primary MPCA sampling method. IBI development data for the Northern Lakes and Forest Ecoregion was collected using the four small plot technique in 2004. The same sampling comparison performed in the Northern Glaciated and Western Cornbelt Plains Ecoregions,

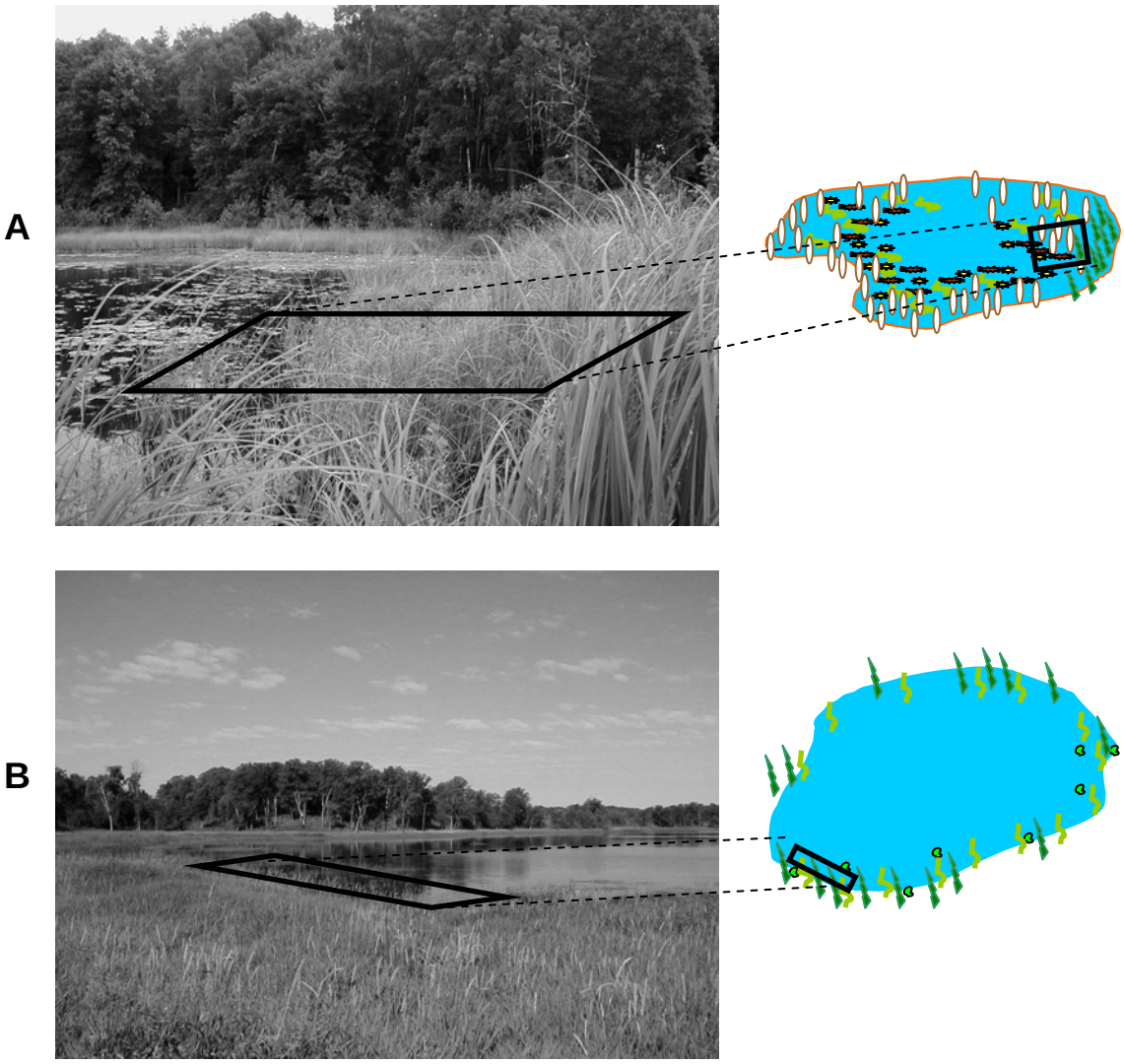

Figure 1. Hypothetical lay-out of a 10 m x 10 m (A) and a 5 m x 20 m (B) plot in two wetlands. In wetland A there is a relatively wide and diverse emergent wetland fringe. Wetland B, on the other hand, has a very narrow emergent fringe. In the diagrams on the right the symbols represent different vegetation communities. In both cases the plots are located at the emergent/aquatic vegetation interface to capture as many of the different vegetation types as possible.

however, did not improve IBI precision (Genet et al. 2006). The MPCA will continue to compare alternate sampling techniques, as we expand and refine wetland biological monitoring.

Ultimately, the sampling plot technique used to develop IBIs in the various Ecoregions of the state needs to be used for consistent application. The single large (100 m^2) plot size should be used for sampling in the North Central Hardwood Forest, Western Corn Belt, and Northern Glaciated Plains Ecoregions (Figure 2). The four small (5 m x 5 m) plot technique should be used in the

Northern Lakes and Forest Ecoregion. Depressional wetland IBIs have not yet been developed or adapted for the Red River Valley, Northern Minnesota Wetlands, and Driftless Area Ecoregions.

Once a representative plot location has been identified choose either the single sample plot or the four small plot sampling technique based on which Ecoregion in the state the wetland occurs (Figure 2). If the single plot technique is to be used, determine which plot shape (square or rectangular) is appropriate. As a general rule, only use the 5 m x 20 m rectangular plot shape when the emergent vegetation fringe is < 5 m wide from the upland boundary to the aquatic vegetation/open water boundary.

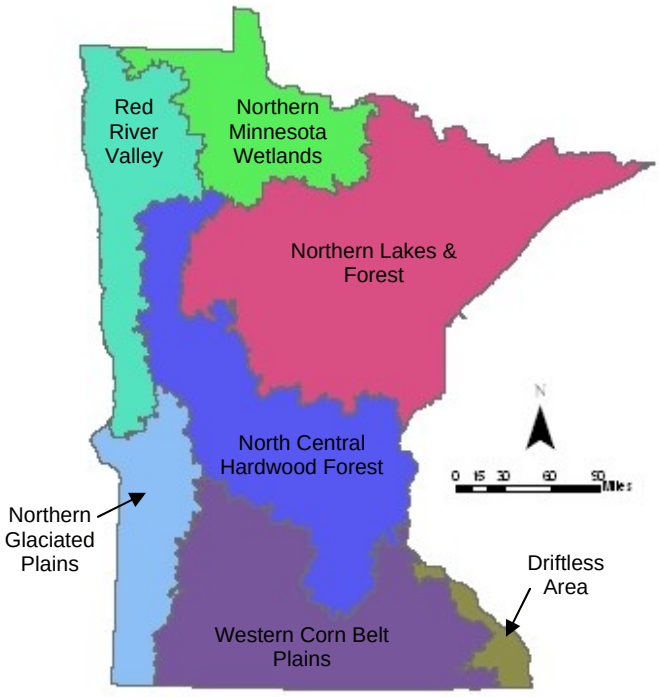

Figure 2. Level III Ecoregions in Minnesota (Omernik 1987). Use the 100 m^2 plot size in the North Central Hardwood Forest, Western Corn Belt Plains, and Northern Glaciated Plains Ecoregions and multiple 25 m^2 plots in the Northern Lakes and Forest Ecoregion.

B.5. Lay-out the plot: To lay-out a plot, first pick a point to be corner #1 and plant one tall gardening stake (Table 1) to mark the corner. Using a tape measure (Table 1), mark off the first side of the plot, according to the dimensions of the determined plot shape, holding the tape measure away from your body and walking outside of the plot area to avoid excessive trampling of the vegetation inside the plot. Stake this point (corner #2). Turn 90 degrees using a compass or best visual judgment, and measure out the second side to corner #3. Repeat these steps, establishing corner #4 and enclosing the plot with four sides. Adjust the corners and sides if necessary. The plot should capture the emergent/aquatic vegetation interface (Figure 1); therefore, a portion of the plot should be in each vegetation type.

B.6. Record releve information: Once a plot, or releve, has been established, begin recording releve level data in the Releve Data Sheet (*attached at the end of this SOP*) or PDA. If using the Releve Data Sheet, establish the **Releve Number** (this is done automatically in the PDA). The releve number consists of the date and

time of the beginning of the releve and should have the following format: month/day/year-hours:minutes:seconds. Record the **Site Name**, **Surveyor's Name**, and **Date**. Determine the **Releve Result,** or use category of the data. A releve is: **Reportable** if the data in that releve will be used for the primary assessment for the site; **Replicate** if the data will be used to determine IBI variance (Genet et al. 2005), for QA/QC purposes, or for secondary assessment; and **Nonreportable** if the data will not be used for any assessment purposes or if the sampling procedures were unable to return a reliable sample. If the **Nonreportable** data use category is selected, document the reason the data should not be used for wetland assessment. Record the **Releve Shape**. If the releve is 5 m x 5 m, also record a **Sample** letter (beginning with A) and **Subsample** number (beginning with 1) for the releve. The **Sample** letter is needed to group multiple 5 m x 5 m plots (i.e., subsamples) together into groups of four. Determine and record the **Average**, **Maximum**, and **Minimum Water Depth** (cm) within the releve. Estimate the percent cover the genus ***Carex*** and **Open Water** occupies in the plot. **Open Water** is defined as standing water that does not have emergent or floating vegetation shading it. Record the approximate position of the releve with a handheld Global Positioning System (GPS) unit. Save the waypoint in the GPS with a file name that consists of the **Site Name** and the **Sample** and **Subsample** indicators (if necessary). If the site was named prior to 2003, use the first six characters of the site name and sample and subsample indicators (if necessary). If the site was named using the year/county/wetland number coding system adopted in 2003 and currently used, record the **GPS File Name** according to the following format: 2 digit year, first four letters of the county, 2 digit wetland number, sample letter (if necessary), and subsample number (if necessary).

Example: the **GPS File Name** for the third 5 m x 5 m plot of the second set of plots (sample) in the site named 04CASS011 should be- 04CASS11B3. If any photographs of the releve are taken, record the appropriate **Photo Info** in the space provided.

B.7. <u>Identify plants within the plot</u>: Next, inventory the plants within the plot. This is done by 'walking the plot' (Figure 3). Begin in corner #1 and walk just inside the plot toward corner #2. Identify and record plants to the lowest

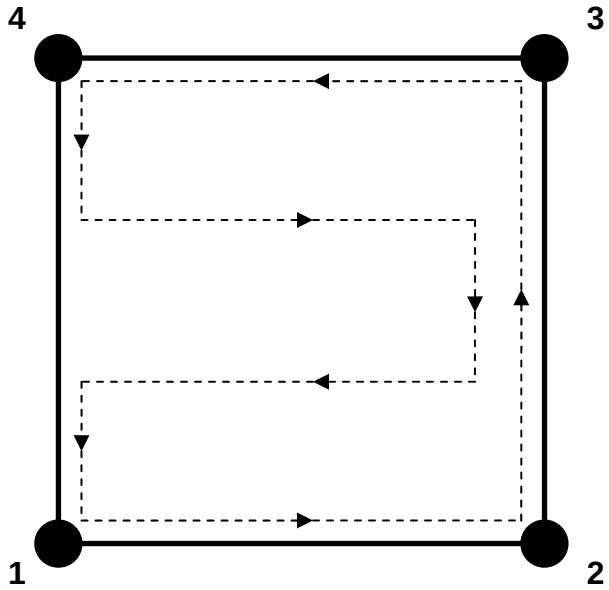

Figure 3. Walking the plot. Begin at corner #1 and follow the arrows until the entire plot has been observed.

taxonomic division possible in the **Species Info** section as you proceed. Continue around sides 2 and 3. After passing corner #4 go about 1/3 of the way of the remaining side of the plot and cut through to the opposite side to observe the vegetation in the interior. Once on the opposite side, proceed down another 1/3 of that side and cut through the plot again. Return to corner #1. In very dense emergent vegetation it may be necessary to do a third interior path to be able to observe the entire plot. For the 5 m x 20 m plot shape, 4-5 interior paths may be necessary to complete the plant inventory.

Record a **Reliability** code (Table 2) for each plant encountered to indicate the level of identification confidence. If there are multiple higher level taxonomic identifications in the same plot belonging to the same group, use the **tsnGroup** space to differentiate individual species (*see B.9*). If a plant is collected to be identified in the laboratory, mark the **Collected** box for that plant.

Table 2. Identification reliability codes.

Reliability Code	Description
7	Unknown
6	cf Genus
5	Genus certain
4	cf species
3	species complex
2	species certain
1	cf var/subsp.
0	variety/subsp. certain

B.8. Estimate cover: For each plant taxa encountered in the plot, estimate the percent cover (proportion of the plot area occupied by the taxa) using the cover class (**CC**) scheme given in Table 3.

B.9. Unknown plants: All plants encountered in the plot should be identified to its lowest taxonomic division possible. When a plant cannot be reliably identified to species in the field, the plant should be recorded using a standard naming convention and be collected for identification later in a laboratory.

The following notation convention should be used to record unknown plants: 1) the scientific name of the lowest known taxonomic division of the plant (e.g., Genus, Family, etc), and 2) a number corresponding to the number of different unknown plants from that taxonomic division encountered in a particular plot. Record the taxonomic division in the **Species Name** column and the number in the **tsnGroup** column.

Example: if one were to encounter an unknown species of the genus *Carex*, *Carex* should be recorded as the **Species Name** and a 1 should be recorded in the **tsnGroup** space. If a different unknown species of *Carex* is encountered in the same plot, the **Species Name** should be recorded as *Carex* and a 2 should be recorded in the **tsnGroup** space.

Table 3. Cover Classes and corresponding ranges of percent cover.

Cover Class (CC)	Percent Cover Range
8	95-100%
7	75-94%
6	50-74%
5	25-49%
4	10-24%
3	5-9%
2	2-4%
1	1%
0.5	0.1-0.9%
0.1	single/few

All unknown plants should be collected, pressed, and dried for positive identification in the laboratory. In the field, collect as much material as necessary, or possible, to facilitate identification of the plant and place in a plastic bag. Label the bag with: 1) the site name, 2) sample and subsample identifiers (if necessary), 3) plant unknown name (i.e., **Species Name** and **tsnGroup** number recorded), 4) date, and 5) collector name. Upon returning to the vehicle, immediately place collection bags into a cooler with ice and keep specimens cool until they can be pressed. Collected specimens must be pressed within 24 hours of collection.

It is unnecessary to collect a specimen for the same unknown taxa in each plot, if multiple plots are to be sampled at the same site, as long as the field crew leader is certain that the taxa is the same species and the naming convention is consistently applied at the site. For example, an unknown species of *Carex* is observed and collected in the first sampling plot. The very same *Carex* is observed in a replicate plot of the same site. It is not necessary to make an additional specimen collection if the crew leader is certain that it is the same species.

C. Data and Equipment Security: Immediately after each day of field sampling, the following actions must be taken to secure the data collected during field sampling and maintain sampling equipment for further use:

C.1. Download Data: Download any and all field data from the PDA, GPS, and digital camera onto the hard drive of the laptop computer. Make an additional copy of these files onto a portable memory source (e.g., 'memory stick', CD) to back up the files. Delete data as necessary on the individual units to reduce duplicate copies of data from downloading the same data multiple times.

C.2. Press Collected Plant Specimens: Specimens must be pressed within 24 hours of collection. Press specimens with a standard plant press that has cardboard ventilators, blotter paper, and newsprint. Each specimen should be placed in an individual piece of newsprint and labeled with the same label as the collecting bag (*see B.9*). Array the plant so that stems and leaves and any flowering or fruiting material are separated and clearly visible. Aquatic plants may require floating in a tray filled with water and arrangement on wax paper.

C.3. Ship Water Chemistry Samples: *See Depressional Wetland Water Chemistry SOP*.

C.4. Equipment Assessment and Maintenance: Assess and maintain sampling equipment as necessary. Clean soiled sediment tubes (*See Depressional Wetland Water Chemistry SOP*). Recharge any flat batteries. Organize, update, and maintain site files and maps. Dry and repair waders as necessary. Acquire fresh ice for cooler.

VIII. QUALITY ASSURANCE AND QUALITY CONTROL

Compliance with this procedure will be maintained through annual internal reviews. Technical personnel will conduct periodic self-checks by comparing their results with other trained personnel. Calibration and maintenance of equipment will be conducted according to the guidelines specified in the manufacturer's manuals.

In addition to adhering to the specific requirements of this sampling protocol and any supplementary site specific procedures, the minimum QA/QC requirements for this activity are as follows:

A. Control of deviations: Deviation shall be sufficiently documented to allow repetition of the activity as performed.

B. QC samples: Ten percent of sites sampled in any given year are re-sampled as a means of determining sampling error and spatial variability.

C. Verification: The field crew leader will conduct periodic reviews of field personnel to ensure that the procedures detailed in this SOP are being followed.

IX. LITERATURE CITED

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MPCA WETLAND VEGETATION VISIT DATA SHEET

Version 2.2 5/2006

Visit Information

Site Name:	Date:	Surveyors Name:	Arrival Time: Leave Time:

Weather Notes:

Site Notes:

Photo Information

Camera Make & Model:	Photo Number(s):	Photo Notes:

Water Chemistry

Field Measurements

	Reportable (A)	Replicate (B)
Temperature (C)	_____	_____
pH	_____	_____
Specific Conductivity	_____	_____
Dissolved Oxygen	_____	_____
Time of Measurement:	_____	_____

Water Grab Samples

Reportable (A)	Replicate (B)		Reportable (A)	Replicate (B)	
☐	☐	Turbidity (1:125 ml)	☐	☐	Calcium, Magnesium (1:500 ml)
☐	☐	Color (1:125 ml)	☐	☐	Preserved with HNO3
☐	☐	Chloride, Sulfate, TOC (1L general)	☐	☐	Nitrogen, Phosphorus (1:250 ml)
			☐	☐	Preserved with H2SO4

Sediment

Reportable (A)	Replicate (B)	
☐	☐	Sediment Sample

Water Chemistry Notes:

Site Stressor Verification

Habitat Alteration

(within wetland & 50 m of surrounding upland)

Checklist:

Wetland	Upland	
☐	☐	Mowing
☐	☐	Excessive herbivory
☐	☐	Shrub removal
☐	☐	Removal of woody debris
☐	☐	Removal of emerg. veg.
☐	☐	Vehicle use
☐	☐	Livestock hooves
☐	☐	Cultivation
☐		Microtopography altered
	☐	Tree plantation
	☐	Tree removal

Sedimentation

Checklist:

- ☐ Sediment deposits/plumes
- ☐ Eroding banks/slopes
- ☐ Turbid water column
- ☐ Soil disturbance in immediate upland (e.g., construction, cultivation)

Hydrologic Modification

Checklist:

- ☐ Ditch *inlet outlet both*
- ☐ Tile Drain # _____ *if multiple*
- ☐ Dredging
- ☐ Weir/Dam Type:_____
- ☐ Grading/Filling (in or near wetland)
- ☐ Stormwater input/culvert #_____*if multiple*
- ☐ Dike, berm or levee _____% of wetland edge
- ☐ Road or RR bed _____% of wetland edge
- ☐ Unnatural connection to other waterbody
- ☐ Source water change
- ☐ Drainage
- ☐ Dewatering in or near wetland
- ☐ Point source (non-stormwater)

Stressor Verification Notes

Sketch of Wetland

MPCA WETLAND VEGETATION RELEVE DATA SHEET

Version 2.2 5/2006

Releve Info

Releve Number:

Releve Result (circle one):
Reportable Replicate Nonreportable

Site Name:

Releve Shape (circle one):
10 x 10 5 x 20 5 x 5

Surveyors Name:

Sample (letter):

Subsample (number; for 5 x 5 plot):

GPS File Name:

Water Depth (cm):
Minimum _____ Average _____ Maximum _____

Photo Info:
Camera Make & Model:
Photo Number(s):

Carex Cover (%): _____ Open Water (%): _____

Releve Notes:

Reliability Code

Code	Description
7	Unknown
6	cf Genus
5	Genus certain
4	cf species
3	species complex
2	species certain
1	cf var/subsp.
0	variety/subsp. certain

Cover Class (CC)

CC	Range
8	95-100%
7	75-94%
6	50-74%
5	25-49%
4	10-24%
3	5-9%
2	2-4%
1	1%
0.5	0.1-0.9%
0.1	single/few

Species Info

Species Name	tsn Group	Reliability	CC	Collected	Notes
				☐	
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				☐	
				☐	
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Species Info

Species Name	tsn Group	Reliability	CC	Collected	Notes
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