

Guidelines for the Standard Collection of Macroinvertebrate Samples from Wadeable Streams v2.0

A. Introduction and Scope

The composition of aquatic macroinvertebrate assemblages can provide information on the ecological condition of streams that may be otherwise difficult to quantify. As the majority of aquatic invertebrates have limited mobility they can be good indicators of local water quality, integrating local and upstream watershed stressors. Additionally, most aquatic macroinvertebrates live from months to years in streams, integrating the effects of multiple environmental stressors over time. Instead of measuring the multitude of possible stressors over different spatial and temporal scales, measuring macroinvertebrate assemblages allows the direct examination of how stressors are impacting biologic integrity.

This standard operating procedure (SOP) document pertains to the collection of macroinvertebrates in wadeable streams for the calculation of the WDNR's macroinvertebrate Index of Biotic Integrity (mIBI, Weigel 2003). For purposes of the mIBI, a wadeable stream is a stream that is wadeable (generally <1.5 meters deep) in greater than 50% of the study reach during base flow. A separate SOP outlines procedures for collecting macroinvertebrates in nonwadeable rivers to calculate the Large River Macroinvertebrate IBI (NW mIBI) (Weigel and Dimick 2011, WDNR 2015).

The Wisconsin Department of Natural Resources uses the mIBI as an indicator of aquatic ecosystem health and to assess against appropriate aquatic life benchmarks. The mIBI was built to reflect structural changes in macroinvertebrate assemblages in response to local and watershed-level disturbance, riparian condition and local habitat quality. As such, the mIBI reflects the response of the macroinvertebrate assemblage to multiple types, and multiple scales, of environmental disturbance (Weigel 2003).

This SOP also pertains to the collection of macroinvertebrate samples to calculate additional metrics useful for describing macroinvertebrate composition besides the mIBI. Additional metrics are calculated by the WDNR Surface Water Integrated Monitoring Systems (SWIMS) database based on the observed macroinvertebrate assemblage. For example, metrics such as the Hilsenhoff Biotic Index, percent Ephemeroptera, Plecoptera and Trichoptera individuals and relative abundance of functional feeding groups, among others, have been used to evaluate the status or trends of particular waterbodies by researchers and resource managers for decades. These metrics can be useful for analyzing macroinvertebrate assemblage composition; the application and relative merit of many of these common macroinvertebrate metrics are discussed in detail in the WDNR's Macroinvertebrate Data Interpretation Manual (WDNR, 2003).

To calculate the mIBI for biologic assessments samples must be collected in a consistent standardized process from standard habitat types. Sections B-F describes the standardized macroinvertebrate data collection methods from wadeable streams. Additional recommended methods for non-standard macroinvertebrate sample collections are discussed in Section G.

B. Summary of Method

Macroinvertebrate sampling for calculation of the mIBI should occur one of two index periods, spring (March-May) or fall (September-November), with the fall index period being heavily preferred. Water levels should be near baseflow. Usually there will not be a long term flow record for that site and staff will have to gauge bankfull height from riparian clues and determine if stream is at or below that level. Samples should be collected in riffle habitats. If no riffles are present, samples may be collected in runs provided there is at least 15% coarse benthic substrate (fine sand or larger, 0.64-2mm size class). Staff should review current data, historic data and field notes from any samples taken in the spring index period or from non-riffle habitats to ensure that the sample is representative of the likely assemblage and that the mIBI should be applied for water quality assessments.

Enter the stream working upstream approaching the target riffle being careful not to disturb the targeted sampling area. Sample the targeted riffle with a D-frame 500 or 600 micron mesh kick net (hereafter “kick net”, see Section G for discussion of alternative mesh size) by holding the net frame firmly against the stream bottom and disturbing the substrate upstream of the kick net with your feet. Dig deeply into the substrate with the heel or toe to dislodge macroinvertebrates from the streambed. Avoid kicking coarse debris into the net such as rocks and woody debris. Make sure that the plume of silt that results from disturbing the substrate is flowing into the net, as this plume also contains the dislodged macroinvertebrates.

After the sample is collected, rinse fine sediment from the net by forcefully swishing the net through the water a few times, being careful not to lose the organisms captured. Removing fine sediment from the net makes laboratory analysis of the sample easier and helps insure adequate preservation of the sample. Discard large sticks, rocks, and leaves from the net after thoroughly rinsing debris to dislodge any clinging macroinvertebrates back into the net.

Transfer the debris and macroinvertebrates to a HDPE or glass wide-mouth jar of sufficient size. Inspect the net and transfer clinging macroinvertebrates into a properly labeled sample jar. The sample debris should occupy less than 1/2 the sample jar’s volume. Initially, preserve the sample with 95% ethanol while in the field. There will be sufficient water in the sample to dilute the final concentration to ~75-80%. Within 48 hours pour-off the alcohol solution and refill with fresh 90-95% ethanol. Samples containing large amounts of organic materials should be preserved and re-preserved several times. Poorly preserved samples result in decayed or brittle organisms and make proper taxonomic identifications difficult or impossible.

C. Safety

Safety precautions of a general nature should be recognized. Collecting samples in extremely hot and cold weather carries the risk of dehydration, heat stroke or hypothermia, respectively. Collection of macroinvertebrates requires contact with stream water and possibly stream sediment. Use precaution (or gloves) when reaching in kick nets to remove large substrates as glass or other sharp objects may be present and difficult to see. All Department-wide safety SOPs apply while collecting macroinvertebrate samples.

Ethanol

Macroinvertebrate samples are preserved in ethanol, with a recommendation that WDNR staff purchase denatured (95%) ethanol as a preservative. Denatured ethanol may be stored and transported without a permit, but is flammable and an eye and skin irritant if handled incorrectly. 95% ethanol has an agent added that makes it unfit for human consumption (i.e. denatured) to prevent the consumption of pure ethanol and subsequent alcohol poisoning. See MSDS for denatured ethanol for complete storage and safety information (<https://tsocorpsite.files.wordpress.com/2014/08/ethanol.pdf>). Pure ethanol (100%) may be used to preserve macroinvertebrate samples as long as target concentration is met (75-85%) as the final preservative concentration in the sample jar and all federal and state permits are obtained and procedures followed. All WDNR manual codes pertaining to the transportation and storage of chemicals must be followed. Due to the relative ease of storage and transportation, denatured ethanol is recommended as the preservative for macroinvertebrate samples.

D. Equipment

- 500-600 micron D-frame Kick Net
- Wide mouth sample jars - 16-32 oz. (500-1000 mL) or larger is recommended
- Ethanol (95%)
- Clear packing tape
- Electrical tape (optional)
- Labels on waterproof paper (two per sample)
- Macroinvertebrate lab slip (printed from SWIMS)
- Pencil
- Tweezers
- Plastic bucket (optional)
- GPS
- Waders

E. Collection Procedures

The following procedures outline the step-by-step approach for collecting macroinvertebrate samples from riffle (always preferred) and run habitats. These procedures assume macroinvertebrate data collection is for the express purpose of calculating the WDNR macroinvertebrate IBI and conducted

during one of the appropriate index periods. Read Section B for a summary of these procedures before following the step by step procedures below. There are many other macroinvertebrate collection procedures that can be used to calculate numerous other macroinvertebrate metrics that do not have step-by-step details outlined in this SOP; some are discussed in Section G.

Sample Collection

In Riffle Habitats

1. Approach the target riffle from downstream or, if approaching from upstream avoid walking over area of riffle that will be sampled.
 - a. Select a portion of the riffle that approximates common riffle characteristics of that stretch of stream. There is no need to develop any type of randomization procedure to select the sample location within or among riffles.
2. Downstream of the target area place the kick net firmly into the stream bottom.
 - a. In riffles with coarse substrate the bottom of the kick net will need to be forcibly placed between cobbles/boulders to ensure dislodged macroinvertebrates cannot escape under the net.
3. In an area approximately one meter long and slightly wider than the kick net use the toe or heel of your wading boot to disturb the stream sediment. Kick deeply to dislodge macroinvertebrates attached to rocks, debris and those in the first few centimeters of sediment.
 - a. You should see a plume of debris (mostly silt) resulting from your kicks, be sure the current is washing this plume into your kick net.
 - b. Continue this process for 1-2 minutes, keeping track of your time.
 - c. Keep track of the total area of stream bed sampled. Following Step 3 this should result in ~0.5 square meters sampled.
4. And the end of the collection period, inspect the kick net and remove any coarse debris.
 - a. While still inside the kick net, large rocks, sticks, macrophytes or leaves should be vigorously rinsed to dislodge any macroinvertebrates and then discarded.
 - b. A debris ball approximately the size of a softball is generally adequate to contain enough macroinvertebrates for identification. You should be able to see larger macroinvertebrates in the debris ball and smaller individuals clinging to the side of the net.
5. If the debris ball appears small or very few individuals are visible in the net repeat the procedures in Steps 1-4 in a new location on the same riffle. Be sure to track the total time collecting and total area sampled among all kicks.
 - a. Do not sample areas that have already been disturbed by walking to your sampling site.
6. If fine sediment is present in the sample debris ball sweep the kick net upstream a number of times to sieve out any silt. Be sure not to lose the contents of the sample while conducting this procedure.

In Run Habitats

7. Some streams will lack riffle habitats, the preferred collection habitat. In these systems the collection should be made in runs, preferably the fastest and shallowest run habitats available with as much coarse substrate as possible in the reach.
8. Downstream of the target area place the kick net firmly into the stream bottom.
9. In an area approximately one meter long and slightly wider than the kick net use the toe or heel of your wading boot to disturb the stream sediment. Kick deeply to dislodge macroinvertebrates attached to rocks, debris and those in the first few centimeters of sediment.
 - a. You should see a plume of debris resulting from your kicks, be sure the current is washing this plume into your kick net.
 - b. If the sample area has particularly sandy substrates try to minimize the amount of sand washing into your net. This can be accomplished by not kicking within the first few centimeters closest to the net allowing sand to settle out while less dense macroinvertebrates are washed into the net.
 - c. Continue this process for 1-2 minutes, keeping track of your time.
 - d. Keep track of the total area of stream bed sampled. Following Step 3 this should result in ~0.5 square meters sampled.
10. And the end of the collection period, inspect the kick net and remove any coarse debris.
 - a. While still inside the kick net, large rocks, sticks, macrophytes and leaves should be vigorously rinsed to dislodge any macroinvertebrates and then discarded.
 - b. A debris ball approximately the size of a softball is generally adequate to contain enough macroinvertebrates for identification. You should be able to see larger macroinvertebrates in the debris ball and smaller individuals clinging to the side of the net.
11. It is common that fewer macroinvertebrates (per unit area) are found in runs than in riffles. If the debris ball appears small or very few individuals are visible in the net repeat the procedures in Steps 7-10 in a new location. Be sure to track the total time collecting and total area sampled among all kicks.
 - a. Do not sample areas that have already been disturbed by walking to your sampling site.
12. If, and only if, very few macroinvertebrates appear present in the sample then target other habitats.
13. Sample by sweeping or jabbing the kick net against woody debris, undercut banks or overhanging vegetation.
 - a. **Important:** record the Habitat Sampled and Composition of Substrate as percentages on the macroinvertebrate lab slip. These data may or may not be used in a macroinvertebrate IBI assessment due to the nature of the sampling.

14. If there is a lot of sand in your sample it may be necessary to conduct some streamside processing before preserving your sample. Excess sand and gravel in the sample jar can grind specimens making them difficult, or impossible to identify.
 - a. Empty the contents of the net into a bucket and add enough stream water to cover. Swirl the contents of the bucket forcibly using your hands.
 - b. The less dense objects, macroinvertebrates and other organic matter, will stay in suspension longer while rocks and sand will settle out quickly.
 - c. Slowly pour the contents of the bucket back through the kick net to sieve out the water. Leave as much sand and gravel in the bucket as possible.
 - d. Repeat this step with new stream water in the bucket as many times as necessary (3 is usually sufficient) until you are sure the vast majority of macroinvertebrates have been separated from the sand and gravel and are back in the kick net.
 - e. Discard sand and gravel back into the stream

Sample Handling and Preservation

15. Streamside, transfer the debris ball into a wide-mouth jar (HDPE or glass). The debris should not fill more than $\frac{1}{2}$ of the space of the sample jar. If the debris ball is larger than $\frac{1}{2}$ the sample jar volume then the sample needs to be split into as many jars as necessary to maintain debris volume at $\frac{1}{2}$ of the space of the sample jar two jars.
 - a. A quart mason jar or 1000 mL HDPE sample container are often used for preservation.
 - b. Remove any large, non-target organisms such as small fish or mussels from your sample if spotted.
16. Be sure to rinse macroinvertebrates from the side of the net into the sample jar. It can be helpful to rinse the net after the debris ball is removed to collect the smaller individuals clinging to the side of the net into a smaller area at the bottom of your kick net. Transfer these individuals to the sample jar.
 - a. It may be difficult to remove some of these individuals from the net. The net can be placed over the mouth of the jar and then “flick” the back of the net to get the more stubborn individuals into the jar.
 - b. Additionally, a small tweezers can be useful for picking macroinvertebrates out of the net. Small, worm-like and long-legged macroinvertebrates may get caught in the net and be difficult to otherwise remove.
 - c. This part of transferring the sample will generally take much longer than Step 15, but is necessary in order to prevent sample bias.
17. Each sample jar needs to have two labels properly identifying the sample. One label needs to be written on waterproof paper and placed inside of the jar with the macroinvertebrate sample. A second label must be securely taped to the outside of the sample jar, preferably on waterproof paper.

18. Each label needs to include:
 - a. Sample ID Number: See Section F
 - b. SWIMS Station ID: See Section F
 - c. Replicate Number: Usually “1”. If two separate samples from same SWIMS Station then identify that by using a Replicate Number.
 - d. Waterbody Name: Official Name
 - e. WBIC: Waterbody Identification Code
 - f. Collector’s Name: Use full surname instead of initials.
 - g. Jar Number: 1 of 1, 1 of 2, etc.
 - h. Project Name: A description that is used to batch samples at the Aquatic Biomonitoring laboratory
19. Preserve the samples to a final target of 75-85% ethanol while in the field.
 - a. Initial preservation can be made with 95% ethanol added directly to the sample. The debris ball will retain some stream water and organic material which will dilute the ethanol concentrations to the final target concentration.
20. Seal the container and invert multiple times to distribute the preservative throughout the sample and store in a secure location. The sample does not need to be stored on ice or refrigerated.
21. Within 48 hours decant the sample using a sieve to prevent loss of organisms, and re-preserve with 90-95% ethanol. There will still be enough water stored in the organic material to dilute the preservative to the final target concentration of 75-85% ethanol. Then seal the sample (with electrical tape) or otherwise indicate on the jar that the sample has been re-preserved and is shelf stable.
 - a. Poorly preserved samples can render a sample unidentifiable, or only able to coarsely identify and result in lost data and inefficient use of staff time.
 - b. Initial ethanol concentrations over 85% can cause macroinvertebrates to become brittle, lose body parts and become unidentifiable to the appropriate taxonomic level. Slightly higher ethanol concentrations are less of a concern for the secondary ethanol preservation once tissues have been fixed by the initial preservation.
 - c. Ethanol concentrations below 75% may be insufficient to preserve the specimens, especially if larger organic matter has not been removed from the sample. Decomposition may result in specimens that become unidentifiable to the appropriate taxonomic level.

F. Data Management

Generating Labslips

All macroinvertebrate samples need to have a macroinvertebrate labslip generated from the SWIMS database (WDNR’s Surface Water Integrated Monitoring System) before they can be submitted to a WDNR approved lab for taxonomic identification. To generate a labslip the user must have the ability to

set up SWIMS project and create a new SWIMS Station ID (if needed). If the user is not familiar with these steps they should contact a SWIMS database manager and receive the necessary training and administrative approvals (i.e. correct roles assigned to your SWIMS username). The preferred method of data management is to set up a SWIMS project, create a new SWIMS Station (only if needed) and print a labslip before sampling begins. For most routine WDNR monitoring the chronological ordering of these steps is appropriate.

In some instances it may be necessary to collect a macroinvertebrate sample before the labslip is generated in SWIMS. For example, if a macroinvertebrate sample is collected as part of a spill response, staff may not know the exact monitoring location before they are on-site. As such, it will not be known if the sample should be related to an existing SWIMS station or if a new one will need to be created. Once the sample is taken staff must create a labslip and re-label the macroinvertebrate jar (inside and outside labels) with the appropriate descriptive information. In these situations it can be helpful to print out a blank labslip to bring in the field to remind yourself to fill out all of the appropriate on-site information. Never send in blank or photocopied SWIMS labslips. Each sampling event needs to have a unique labslip with a unique database key for tracking and reporting purposes.

Filling Out Labslips

In SWIMS

- a. Form – Wadeable Macroinvertebrate Field Data Report (3200-081)
- b. Project – Selected from existing SWIMS project
- c. Data Collectors – Staff names must be in SWIMS database
- d. Station - Selected from existing SWIMS station
 - a. If you need to create a new station this must be done before generating the labslip
 - b. Exact location (GPS) can be included later in the site information block of the field visit form. Also include approximate distance upstream or downstream of the road crossing (or SWIMS Station ID) if the sample isn't exactly at the SWIMS station location, usually located at a road crossing. Use this feature instead of creating a new SWIMS station to avoid having many clustered stations on the same stretch of stream.
- e. Start Date – Day of sample
- f. End Date – Optional
- g. Time – Optional
- h. Account Code – Code tied to SWIMS project
- i. Program Code - WY for Water Quality, or other as appropriate
- j. Report to User ID and Report to Name – Name of staff ultimately responsible for interpreting results, typically the local Water Quality Biologist.
- k. Report to Address – Optional
- l. Field Sample ID – Assigned of the form: Year Month Day – County Code – Field Number (number of samples that day in that county).

- i. Written as YYYYMMDD-CY-FN
- ii. 20160603-38-02 is the second sample taken in Marinette County on June 3rd 2016.
- iii. County codes are listed in Appendix A
- iv. Each labslip also has a unique Database Key when the labslip is generated; optionally, this can be written on the jar labels if there is confusion over the Sample ID. DO NOT copy labslips for use in multiple samples.
- m. Select labslip parameters button
 - i. Sampling Device -D-frame Kicknet is required for standard macroinvertebrate IBI calculations.
 - ii. Reason for Sampling – Optional
 - iii. Latitude/Longitude – Check “Use Location Data from Station” unless another coordinate is required (e.g. nearest riffle and sample location is >50m from SWIMS station but still want the sample attached to SWIMS station at the road crossing)

In the Field – Page 1

- n. Habitat Sampled – most often “Riffle” or “Run”.
- o. Total Sampling Time and Estimated Area Sampled – add together if more than one “kick” was used.
- p. Number of Samples in Composite – The number of individual “kicks” were used in the sample
- q. Replicate No – for one sample, record 1 of 1, otherwise 1 of 2 and 2 of 2, etc. Print a new labslip for each replicate.
- r. Water quality measurements – Optional
 - a. Includes Water Temp, D.O., pH, Conductivity, Transparency, Measured Velocity
- s. Stream Characteristics – Required
 - a. Includes Water Color, Estimated Stream Velocity, Average Stream Depth and Average Stream Width
 - i. Do your best to estimate these conditions, it may prove useful years later if very little other water quality information is available for this particular site
- t. Composition of Substrate Sampled
- u. Embeddedness of Substrate
- v. Canopy cover at Sample Site
 - i. For all Steps t, u and v record what you actually sampled, not an average of the reach.
 - ii. Round to the nearest 10%

In the Field – Page 2

The Stream and Watershed Descriptors on page 2 of the macroinvertebrate labslip are meant to qualitatively describe environmental stressors to the aquatic community that may not be readily identifiable through basic monitoring parameters. Unless the collector(s) have spent a

significant amount of time monitoring and travelling throughout the watershed it is anticipated that most of the answers to specific stressors will be Uncertain (U). For the most part, only the Physical and Sources of Stream Impacts parameters will be readily identifiable on-site. There may be cases where Biological or Chemical stressors are exceptionally severe and obvious on-site, but these are expected to be typically recorded as Uncertain (U). Filling out this side of the form is optional, but recommended if stressors are apparent to the sampler.

- w. Fill in each possible category with the following codes, also listed on the lab slip:
- i. N = Not a problem. The collector(s) determine that these stressors are very likely not affecting the aquatic community.
 - ii. U = Uncertain. The impacts of these stressors are unknown. If the cell is left blank it will be assumed as Uncertain. If all cells are left blank the page 2, then these results will be considered incomplete and not recorded.
 - iii. PL = Present Low Impact. These stressors are present but likely not in a great enough extent or severity to impact the aquatic community.
 - iv. PH = Present High Impact. These stressors are present in a great enough extent or severity to likely impact the aquatic community.

Chain of Custody Samples

Macroinvertebrate samples are sometimes used in enforcement cases or other situations where it is necessary to maintain a chain of custody (CoC). All WDNR policies and forms pertaining to water chemistry CoC must be followed when collecting and storing macroinvertebrate samples needing a CoC (<http://intranet.dnr.state.wi.us/int/es/science/ls/Forms/COC.htm> and http://intranet.dnr.state.wi.us/int/aw/air/Compliance/IntranetSCETFinal/FieldDocuments%5C4100145_fill.pdf). These links are active as of the date this SOP was finalized, however they may change in the future and staff will need to keep up to date with all WDNR procedures regarding CoC.

After collection of the macroinvertebrate sample the sample jar should be sealed and taped shut with nylon-reinforced tape. Fill out all labeling information on macroinvertebrate sample jar, macroinvertebrate lab slip and CoC form (see website above). The collector(s) name must be on the CoC form and remain in possession of the sample unless an official transfer of custody occurs. If necessary, samples should be stored at a WDNR office in a locking cabinet until sample delivery or shipment. If the sample is opened to decant ethanol and re-preserve, then the identified custodian needs to re-tape the jar and record opening of the sample on the CoC form. Two staff members should be present for this activity and certify (by signing the CoC form) that the sample was opened, decanted, re-preserved and re-sealed following the Macroinvertebrate SOP. If the sample is delivered immediately it will not be necessary to replace the ethanol in sample jar. When the sample is transferred to the taxonomist the CoC form needs to be updated and a receipt of transfer obtained from the taxonomic laboratory (this can be specified as “received” on a photocopy the CoC form).

G. Non-Standard Collection Procedures

For a number of reasons the specific habitat requirements required for application of the macroinvertebrate IBI may not be present at a given site. Alternative sampling methods can be used to assess the current biologic status of macroinvertebrates at these sites, but it is not recommended that the macroinvertebrate IBI be applied. However, other metrics that describe macroinvertebrate structure may be a better estimation of the current condition of macroinvertebrates at that site, depending on the specific project objectives. The following briefly describes situations where staff may want to consider alternative macroinvertebrate collection methods; this list is not exhaustive of all possible scenarios and procedures.

Mesh Size

Standard mesh size for kick nets are important to ensure the individuals in the sample are not selectively biased towards smaller or larger individuals than were collected during mIBI development. This is especially important for small individuals such as Chironomidae, among others, that may pass through larger mesh openings. WDNR requires a 500-600 micron mesh net for application of the mIBI in water quality assessments. However, anecdotal evidence suggests that historically some nets sold as 500-600 micron mesh size actually had much larger openings. While this problem appears to be resolved (based on current measurements from one popular brand), if larger mesh sizes are used this needs to be documented in the comments section on the lab slip. The data will need to be verified that it is still appropriate for mIBI calculation (e.g. is percent Chironomidae individuals within acceptable ranges compared to similar systems?).

Streams Lacking Coarse Substrate

There are many streams that will not have riffle or run habitats with >15% coarse habitat available for sampling using the standard procedures. Macroinvertebrate data may still be desired at these sites to inventory current species presence/absence, track changes in species composition or metrics over time, detect changes before-after stream restorations, and evaluate the recovery from a spill or other disturbance, among others. Macroinvertebrate data can still be useful for these projects as long as 1) appropriate metrics are selected to describe the assemblage based on the project objective 2) consistent sampling techniques applied over the timespan of the project when analyzing trends and 3) all sampling methods and project objectives are recorded in a SWIMS project and associated with each sample event.

Options for sampling soft bottomed streams

- 1) Collect multi-habitat samples. The benthos of soft-bottomed streams is usually less productive than that of coarse riffle habitat in terms of macroinvertebrate abundance. The most productive areas of these streams may be coarse woody debris and overhanging vegetation. Samples can be collected by specifically targeting coarse woody debris, overhanging vegetation, macrophytes

and kicks or sweeps of the soft sediment. Be sure to select “Proportionally-Sampled Habitat” and estimate the Composition of Substrate Sample on the macroinvertebrate lab slip.

- 2) Sample coarse material near bridges or culverts. These materials are artificially placed, but if the rocks are of sufficient size and in a riffle standard sample collection techniques and standard assessments may still apply. If the coarse sediment is not in a riffle, or is too large to disturb and generate a good sample, then this can be included as part of a multi-habitat sample and alternative metrics used to evaluate the assemblage.
- 3) Hester-Dendy or other artificial samplers. WDNR standard protocols for nonwadeable river macroinvertebrate collections require the deployment of Hester-Dendy samplers. In large, but still wadeable streams, with shifting sand substrates it may be possible to deploy Hester-Dendy samplers to collect information on the macroinvertebrate assemblage. Additionally, there are historic records of macroinvertebrate data in wadeable streams collected by WDNR staff. If a project requires comparison to historic observed data, deployments of Hester-Dendy or “rock-baskets” may be the most appropriate sampling method.

Species Catalogs or Detection of Rare Species

- 4) If a project objective requires a catalogue of all of the species likely present in the ecosystem, or is specifically targeting the detection of rare or sensitive species, then it may be appropriate to use a multi-habitat sampling strategy (See Section G.1, above). Even if there is suitable riffle habitat a multi-habitat sample will increase the chance of detecting rare species, or species not suited to fast-water habitats. If this is the project objective, discuss this with the taxonomist beforehand as it may be appropriate to have an entire sample processed instead of a random subsample.

H. SOP Tracking and Updates

Version Number	Date	Sections	Name	Approval
1.0	06/01/2000	All	Miller	
2.0	11/10/2017	All	Stream Tech Team	Mike Shupryt

I. References

Weigel, B.M. 2003. Development of stream macroinvertebrate models that predict watershed and local stressors in Wisconsin. *Journal of the North American Society*. 22(1):123-142.

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WDNR 2015. Large river macroinvertebrate sampling SOP, v2.0. Wisconsin Department of Natural Resources, Bureau of Water Quality. Madison, WI. PUB-WY-080-2015.

This document is available electronically on the WDNR's website.

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EGAD # 3200-2017-35

Appendix A

County Codes:

Adams (01)	Iowa (25)	Polk (49)
Ashland (02)	Iron (26)	Portage (50)
Barron (03)	Jackson (27)	Price (51)
Bayfield (04)	Jefferson (28)	Racine (52)
Brown (05)	Juneau (29)	Richland (53)
Buffalo (06)	Kenosha (30)	Rock (54)
Burnett (07)	Kewaunee (31)	Rusk (55)
Calumet (08)	La Crosse (32)	St. Croix (56)
Chippewa (09)	Lafayette (33)	Sauk (57)
Clark (10)	Langlade (34)	Sawyer (58)
Columbia (11)	Lincoln (35)	Shawano (59)
Crawford (12)	Manitowoc (36)	Sheboygan (60)
Dane (13)	Marathon (37)	Taylor (61)
Dodge (14)	Marinette (38)	Trempealeau (62)
Door (15)	Marquette (39)	Vernon (63)
Douglas (16)	Menominee (40)	Vilas (64)
Dunn (17)	Milwaukee (41)	Walworth (65)
Eau Claire (18)	Monroe (42)	Washburn (66)
Florence (19)	Oconto (43)	Washington (67)
Fond Du Lac (20)	Oneida (44)	Waukesha (68)
Forest (21)	Outagamie (45)	Waupaca (69)
Grant (22)	Ozaukee (46)	Waushara (70)
Green (23)	Pepin (47)	Winnebago (71)
Green Lake (24)	Pierce (48)	Wood (72)