

Lower Nemadji River Biological and Water Quality Monitoring

Quality Assurance Project Plan

EPA Grant Funding Source: GLRI Grant titled: Addressing excess sedimentation impairments in the Nemadji River Basin (Remedial Action Plan Project 6-5)

Grant #: GL-00E01474 sub 1a

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Prepared: Molly Wick, 08/27/2015

Revision #:

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1 Sept. 2015 *

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* Authorization for field work given in May 2015
Standard DNR monitoring protocols used

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Distribution List

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Executive Summary

The Nemadji River was included in the St. Louis River Area of Concern due to sedimentation issues associated with the Excessive Loading of Sediment and Nutrients Beneficial Use Impairment (BUI). In order to assess the influence that sediment loading in the Lower Nemadji River has on the natural communities, WDNR staff will collect fish and macroinvertebrate data along the Lower Nemadji River. Fish and macroinvertebrate community survey data will be used to calculate Indices of Biological Integrity (IBIs) and assess community health in the Lower Nemadji River. Water quality data will also be collected in the Lower Nemadji River to identify potential stressors to IBIs.

A. Project Organization

Molly Wick, Project Coordinator, WDNR

- Project and QAPP coordination
- Assist with data collection as necessary

Craig Roesler, Principle Investigator, WDNR

- Coordinate and lead macroinvertebrate and water sampling
- Coordinate laboratory analysis
- Assure laboratory data quality on reported data

Paul Piszczek, Fish Biologist, WDNR

- Coordinate and lead fish surveys

Donalea Dinsmore, Quality Assurance Coordinator

- Review and approved Quality Assurance Project Plan
- Perform project audits as necessary

Kurt Schmude, Associate Professor, University of Wisconsin Superior (UWS)

- Aquatic Invertebrate Identification

Wisconsin State Lab of Hygiene

- Water Quality Sample Analysis

University of Wisconsin (UWS) Laboratory

- E. coli Sample Analysis

Additional field data collection assistants include but are not limited to the following WDNR staff:

- Rachel Preacher
- Aaron Nelson
- Kirk Olson
- Madeleine Wedge

Problem Definition/Background

The Nemadji River was included in the St. Louis River Area of Concern due to sedimentation issues associated with the Excessive Loading of Sediment and Nutrients Beneficial Use Impairment (BUI). This watershed is situated in the extremely erodible soils of the Lake Superior Clay Plain, and is therefore very susceptible to erosion. A history of logging and agriculture in the watershed has further exacerbated erosion. The Nemadji River is listed as impaired for turbidity according to section 303d of the EPA's Clean Water Act in Wisconsin and Minnesota.

Previous data collected by the WDNR on the Nemadji River upstream from CTHC suggest that the Nemadji hosts healthy biological communities. Based on indices of biological integrity (IBIs), all the sites sampled previously had good or excellent fish and macroinvertebrate communities, with the exception of a site on Crawford Creek, which is downstream of a site contaminated with creosote, PAHs and dioxins (Roesler, 2014). While assessments upstream of CTHC in Wisconsin suggest that the river hosts healthy biological communities, the Lower Nemadji represents a significant data gap for biological and water quality monitoring:

- Previously the most downstream macroinvertebrate sample from the Nemadji River was collected at CTH W, 31.2 miles above the mouth.
- Previous fish surveys on the Lower Nemadji include boom and barge electrofishing surveys collected at three locations downstream from CTH C, 11.9 miles from the mouth (August 1962), and at one location approximately one-mile upstream from CTH C (May 1952).
- The most downstream water chemistry data on the Nemadji River was collected at CTH C, 11.9 mi above the Nemadji River mouth.

The lower reaches might be expected to be most degraded due to the cumulative impacts upstream and the urban influence of the city of Superior, Wisconsin, which is located at the mouth of the Nemadji. To help determine if sedimentation in the watershed has had an impact on biota in the system, we will collect macroinvertebrate, fish, and water quality data on the Lower Nemadji. Biological community data will be used to calculate IBIs and determine if the Lower Nemadji River healthy or impaired biological communities. Water quality data will be collected to identify potential stressors for biota impairments.

This project is part of a more extensive assessment of the Excessive Loading of Sediment and Nutrients Beneficial Use Impairment in the Nemadji watershed, which also includes modeling historic sediment loading, and calculating a modern sediment load for comparison to a previous calculation based on 1970's data (NRCS, 1998).

Project Location

All biological and water quality monitoring work is focused on the Lower Nemadji, which would be expected to be the most impacted due to its location in the urban area of Superior, Wisconsin. For the purposes of this project, Lower Nemadji is defined as the reaches downstream from Finn Road, which is 19.9 miles above the mouth. Figure 1 shows the Lower Nemadji project area.

Project Data Quality Objectives

DQO 1 Evaluate the health of the biological communities in the Lower Nemadji River through the use of indices of biological integrity (IBIs).

- Macroinvertebrate IBI – In order to calculate an IBI for the macroinvertebrate community, macroinvertebrate data is needed along the lower reaches of the Nemadji River.
- Fish IBI – In order to calculate an IBI for the fish community, fish surveys are needed along the lower reaches of the Nemadji River.

DQO 2 If impairments are observed, connect any observed impairments in biological community health to observed and documented water quality stressors.

- In order to connect any possible community health impairments to water quality stressors, water quality data is needed along the lower reaches of the Nemadji River from the same season as the macroinvertebrate and fish data are collected.

The main data user for the data generated will be water resource managers within the DNR. Data representativeness, comparability, completeness, precision, and accuracy are each a part of the data quality objectives that must be considered during the project planning stages and during data assessment.

Data Representativeness

Representativeness expresses the degree to which sample data accurately represent the site, specific matrices, and parameter variations at a sampling point. Representativeness is a qualitative parameter which is dependent on both the proper design of the sampling program, proper selection of laboratory methods, and stability of the laboratory methods. The representativeness criterion is best satisfied by making certain that the sampling locations, procedures and quantities are selected based on the project objectives, and that suitable analytical procedures are utilized, preservation requirements are met and holding times are not exceeded in the laboratory.

Sampling sites will be chosen (as described in more detail below) to be representative of the main stream of the Lower Nemadji River. For macroinvertebrate sampling, the sites and sampling methods were chosen in order to include all habitat types present in the Lower Nemadji River. For fish surveys, sampling sites will be chosen to include all habitat types in the Lower Nemadji, stratified across the range of land use types in the lower Nemadji watershed. The land use (forest, urban, agriculture) could influence the quality of in-stream habitat, so to get a representative sample of fish in the river, sample sites will be stratified across all types of land use.

Water quality samples aim to represent average water quality of the main stem of the Lower Nemadji River. The water quality samples will be taken from the middle of the river using a Kemmerer sampler, in order to ensure well-mixed water that is representative of the lower main stem of the river. Samples will be taken below the surface to avoid influences from surface water. Studies have shown that using a modified-random sampling design based on stream-crossing road access sites results in no statistically significant difference in results compared to a true random sampling design (Miller et al., 2009). Field duplicates can be used to ascertain the variance of the data as generated by both field and laboratory protocols. During water quality sampling, two duplicate samples will be collected.

Field duplicate analyses at a minimum rate of 10% of samples can be used for macroinvertebrate and water quality sampling to ascertain the variance of the data as generated by both field and laboratory protocols. For fish surveys, a range of capture techniques will be used in order to ensure that a representative population of fish is sampled.

Comparability

Comparability expresses the confidence with which one data set can be compared with another. Comparability is defined as similarity of chemical or physical results from different batches of samples or different sampling events. Sample data may be compared with other measurement data if consistent documented analytical procedures are used for similar samples and sampling methods and conditions.

In order to ensure comparability with macroinvertebrate and water quality samples taken from the Upper Nemadji and on similar rivers throughout the state, WDNR standard sampling protocols will be used, along with standard preservation, and shipping methods. For water sample analysis, the chemical parameters were chosen to be comparable to long term trend monitoring sites. Analysis for dissolved reactive phosphorus (lab filtered orthophosphate) provides the opportunity to understand biologically active phosphorous. The analysis of certified reference materials as conducted according to standard operating procedures of the Wisconsin State Lab of Hygiene will be used to provide data on comparability.

Because of the difference in data collection methods as well as a significant difference in ecological conditions present in the Lower Nemadji compared to the lower St. Louis River estuary, the data collected through this effort will not be included in the SLRAOC Benthic Macroinvertebrate index.

Completeness

Completeness is defined as the percentage of measurements or amount of data required in order to make water quality management decisions. The completeness goal is that all data necessary to meet the two data quality objectives above is collected. Completeness of the deliverable will be measured for each set of data received by dividing the number of valid (passing QA/QC requirements) measurements actually obtained by the number of measurements made. Completeness has been set at 90 %. This empirical determination of completeness still does not answer the question as to whether or not all data necessary will be generated for a valid assessment of community health (DQO 1) and identification of potential links to water quality (DQO 2). Field sampling according to DNR and laboratory SOPs following standard protocols with ten percent field duplicates and QC checks will suffice to yield enough information to make decisions to meet the objectives above. The study was designed, as described in more detail below under Sample Process Design and Sampling Methods, to collect all the necessary data to meet objectives. Enough data should be collected such that an assessment of biological community health in the Lower Nemadji River can be made. For macroinvertebrates, six kick samples and at least 3 vegetation/debris samples will be collected between Finn Road and the mouth of the Nemadji, a distance of 19.9 miles. Therefore the sample density will be approximately one sample every 2 miles. For fish surveys, five one-mile segments will be surveyed within the 19.9 mile project area, so approximately $\frac{1}{4}$ of the project area will be surveyed. For water quality surveys, three sites will be sampled within the project area, which is a density of one site per 6.6 miles.

Macroinvertebrate sampling will be conducted in the fall. Surveys will be done in the fall because lower water levels result in easier access, but also because the populations sampled may be less influenced by short-term spring flood events and more representative of the river. Water quality sampling will be conducted from May through October to ensure that dataset representative of the complete growing season is obtained.

Precision

Precision is a measure of the degree to which two or more measurements are in agreement. Precision measures the reproducibility of measurements under a given set of conditions. Field precision (variance) is assessed through the collection and measurement of field replicates at a rate of approximately 10%.

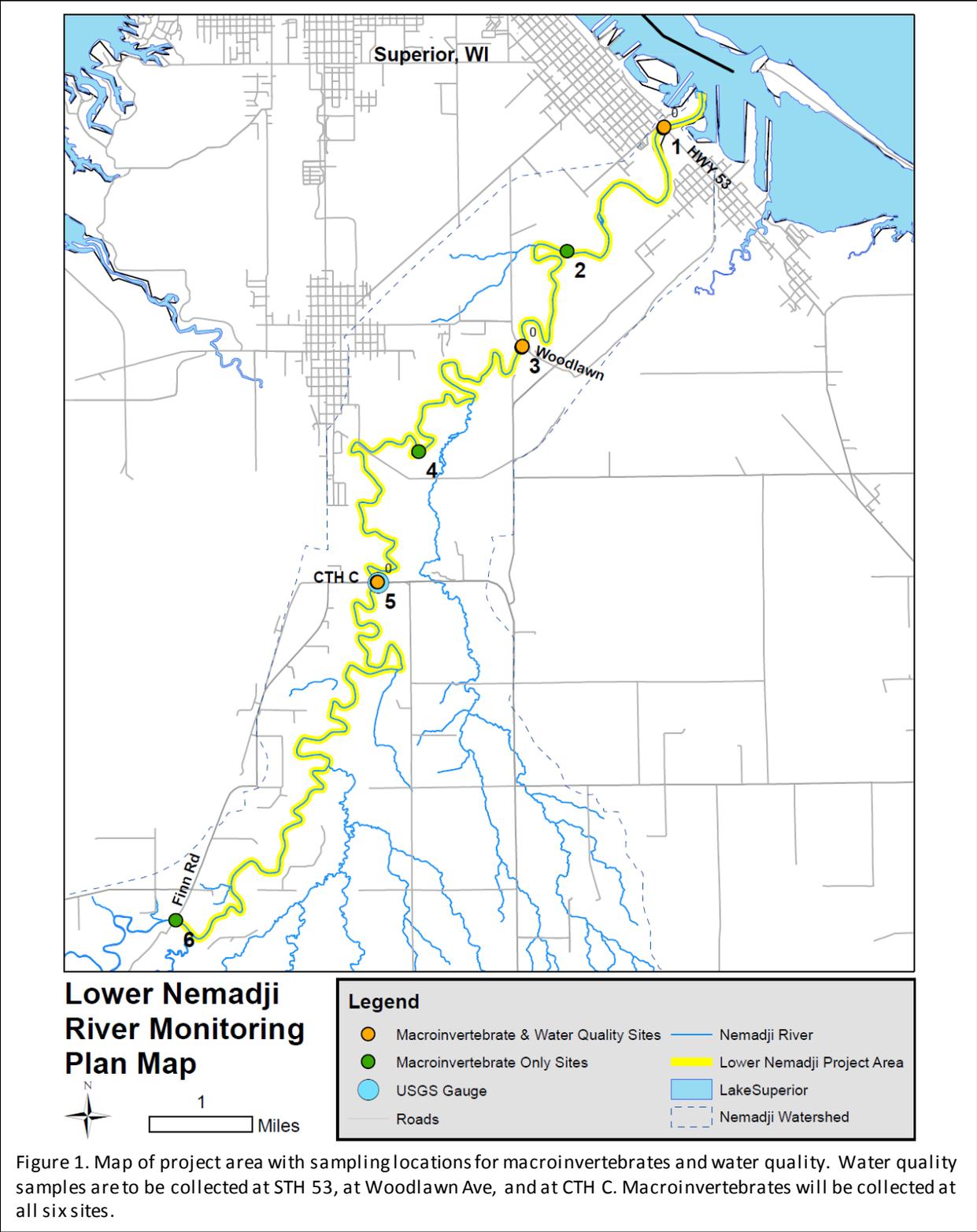
The duplicate samples will be used to estimate sampling and laboratory analysis precision. Two duplicate kick samples will be collected for macroinvertebrates. Results for any duplicate samples with IBI scores that vary beyond 25 points this will be evaluated by internal macroinvertebrate experts to determine if some corrective action is necessary. Field conditions and other metrics may be included in this evaluation (e.g. species richness). During water quality sampling, two duplicate samples will be collected. Table 2 below shows the detection limits that will be applied to the analyses.

Accuracy

Accuracy is the degree of agreement between an observed value and an accepted reference value. Accuracy measures the bias of the measurement system. Sources of this error are the sampling process, field contamination, preservation, handling, sample matrix, sample preparation, and analysis. Accuracy in the field is assessed through the adherence to all sample handling, preservation, and holding times.

During water quality sampling, two field blanks of deionized water will be collected and treated as a sample. The field blank is used to identify errors or contamination in sample collection and analysis. The WSLOH has standard procedures for instrument calibration quality control, which are found in the *Quality Assurance Manual - Environmental Health Division - Wisconsin State Laboratory of Hygiene*. Table 2 below shows the control limits that will be applied to the analyses.

The GPS used to locate and record site locations will have an accuracy of approximately 10m.



Task Description and Schedule

More detailed information on specific tasks can be found below under Sample Process Design.

Macroinvertebrates:

September - October 2015:

- Collect 6 standard kick samples on Lower Nemadji River + duplicate samples at 2 sites; samples will be collected from woody debris since gravel/cobble substrate is not present at most sites.
- Send samples to Dr. Kurt Schmude's lab at University of Wisconsin-Superior (UWS) for identification to lowest possible taxonomic level

Winter 2015 – 2016:

- Receive results from macroinvertebrate sampling
- Calculate macroinvertebrate IBIs and interpret results

Fish

July/August 2015:

- Electrofishing surveys at 5 one-mile segments on Lower Nemadji
- Targeted minnow traps and fyke and gill net surveys on Lower Nemadji
- Enter and summarize data

Winter 2015 – 2016:

- Calculate fish IBIs and interpret results

Water Chemistry

May – October 2015:

- Collect one sample per month at each of three sites on Lower Nemadji River and send to Wisconsin State Lab of Hygiene and UWS Laboratory for analysis

Winter 2015 – 2016:

- Review and summarize water quality data and interpret results

Personnel, Special Training Requirements or Certifications

Prior to any work-related effort, all WDNR staff will be trained in sampling techniques by experienced personnel, and will sample or process samples only under the supervision of personnel experienced in these sampling techniques until they demonstrate proficiency. All project personnel will also have met program safety training requirements.

Kurt Schmude, University of Wisconsin Superior, has 28 years of experience in aquatic invertebrate biology and aquatic invertebrate taxonomy.

Wisconsin State Lab of Hygiene is a Wisconsin certified lab and provides primary analytical laboratory support to WDNR.

University of Wisconsin Superior Laboratory is a Wisconsin certified laboratory for E. coli analysis.

Documentation and Records/Data Management

Field data for macroinvertebrate sampling will be recorded on standard field sheets found in *WDNR Guidelines for Collecting Macroinvertebrate Samples from Wadable Streams*. Macroinvertebrate samples will be sent to Dr. Kurt Schmude's lab at University of Wisconsin-Superior for identification. Results from the macroinvertebrate sampling will be provided in excel spreadsheet format to WDNR. Results will then be stored on WDNR computers and results will be entered into SWIMS.

Field data for fish community surveys will be recorded on standard field sheets. Data will then be entered into excel spreadsheets and stored on WDNR computers. Fish data will also be entered into the WDNR's SWIMS database.

Field data for water quality sampling will be collected on standard lab slips. WSLOH staff will enter water quality analyses into the SWIMS database as part of their routine duties.

The PI will review the data for potential outliers and other possible problems and interpret the data (see Reporting below). One final report summarizing results will be produced by the PI.

Macroinvertebrate, fish, and water quality data can be highly influenced by local and temporal environmental factors. The results of this project will be representative of the conditions of the Lower Nemadji River during the time of sampling (2015 field season).

B. Measurement/Data Acquisition

Sample Process Design (Experimental Design)

Macroinvertebrates

Macroinvertebrate samples will be collected from sample sites between Finn Road (19.9 miles above the mouth) and the Nemadji River mouth (See Figure 1 and Table A) in September - October 2015. Sample sites may be adjusted in the field based on access and field observations. Any changes to sample site will be noted in field notes. At two of the sites, duplicate kick samples will be taken. Kick samples are the most appropriate sampling method for macroinvertebrates due to the flooding potential of the Lower Nemadji River. Detailed sampling methods are provided below under Sampling Methods.

Macroinvertebrate samples will be prepared in the field and delivered to Dr. Kurt Schumde's lab at UWS. Macroinvertebrate data will be used to calculate biotic indices to evaluate the health of the system and assess impairments to macroinvertebrate populations in the Lower Nemadji.

Table A. Approximate macroinvertebrate sampling locations.			
Site No.	Name	Latitude	Longitude
1	STH 2/53	46.6966	-92.0349
2	(half way between adjacent sites)	46.6794	-92.0549
3	Woodlawn Road	46.6660	-92.0644
4	(half way between adjacent sites)	46.6515	-92.0856
5	CTH C	46.6332	-92.0944
6	Finn Road	46.5860	-92.1362

Fish

Fish surveys will be conducted in order to identify the assemblages of fish present in the Lower Nemadji during the mid-summer months. Surveys will be conducted during a consecutive three to four-day period in July or August 2015. Surveys will be conducted in a variety of physical aquatic habitat types at five, one-mile reaches along the lower Nemadji. Locations of sample reaches will be chosen in July 2015 during a reconnaissance outing of the Lower Nemadji. Survey segments will be chosen based on accessibility and will include habitats representative of the Lower Nemadji River stratified across the range of land uses along the Lower Nemadji (urban, forest, hay/pasture). The land use (forest, urban, agriculture) could influence the quality of in-stream habitat, so to get a representative sample of fish in the river, sample sites will be stratified across all types of land use. The intention is to choose a

combination of reaches that together are representative of the Lower Nemadji as a whole, and result in a representative sample of fish in the Lower Nemadji.

The limited previous surveys on the Lower Nemadji noted limitations in electrofishing effectiveness due to fairly deep and turbid water; many fish escaped capture. In order to ensure that the surveys produce results representative of the entire fish community present, both passive and active fish sampling methods will be used. Electrofishing will be conducted at all of the sites, and gill nets, minnow traps, and mini-fyke nets will be available to use as well. Selection of the most appropriate gear type(s) for a particular target segment will be at the discretion of the experienced on-site fisheries biologist. Detailed sampling methods are provided below under Sampling Methods.

Fisheries staff will count, identify, gender, and measure fish length in the field. Following data entry, a data summary will be shared with the PI following data collection. Fish survey data will be used to calculate established fish biotic indices to evaluate the health of the system and assess impairments to fish populations in the Lower Nemadji following data collection.

Water Chemistry

One sample will be collected each month from May to October, 2015 at three sites on the lower Nemadji River. Samples will be collected on the second Wednesday of each month. The objective is to understand the average water quality conditions in the Lower Nemadji River throughout the growing season. Sample sites will be located at State Hwy 53 (0.7 miles above mouth), at Woodlawn Rd (5.4 miles above mouth), and at CTH C (11.9 miles above mouth). Sample sites were chosen to be representative of average water quality in the main stem Nemadji River as well as based on access to the river. CTH C was chosen based on access, because of previous sampling that has occurred at that site, and because of the existence of a USGS staff gage at the site (USGS Gage 04024430). Sampling sites were not chosen to be located at tributary confluence points, as the project objective is to evaluate average water quality in the main stem, not to evaluate tributary contributions or influences. The samples will be collected from bridges in the middle of the river below the surface of the water, such that well-mixed water representative of the Lower Nemadji River is collected and analyzed. Therefore it is not necessary to co-locate these samples with biotic surveys and still be able to compare results of biotic surveys with water quality results.

These samples will be analyzed in the field for dissolved oxygen, pH, temperature, and conductivity using a water quality probe, and transparency using a transparency tube. Samples will be analyzed by the Wisconsin State Lab of Hygiene (WSLOH) for the following parameters:

- Total phosphorus (TP)
- Orthophosphate (OP)
- Total Kjeldahl Nitrogen (TKN)
- Nitrate plus Nitrite (NO₃+NO₂)
- Ammonia (NH₄)
- Total Suspended Solids (TSS)
- Turbidity

Grab samples will also be analyzed for E. coli. Preliminary samples collected at the sample sites yielded results in excess of 1000 cfus, significantly above the pre-2012 single sample maximum criteria of 235 cfus. The 2012 WISCALM criteria for E. coli state a geometric mean threshold of 126 cfus. The EPA criteria for impairment is that 90% of the samples must be below the statistical threshold value of 410

cfus. The presence of E. coli bacteria can be an indicator that there is a source of wastewater or other contaminants to the stream, which could impact fish and macroinvertebrate community health. Samples will be analyzed for E. coli by UWS Laboratory. Detailed sampling methods are provided below under Sampling Methods.

Sampling Methods and Sample Holding

All boats, equipment and gear used in waterbodies outside of the Nemadji River shall be decontaminated according to WDNR protocols prior to and following use in the Nemadji River (Manual Code 9183.1 Boat, Gear, and Equipment Decontamination Protocols; WDNR Best Management Practices for Boat, Gear, and Equipment Decontamination, April 2015).

Macroinvertebrates

Six standard kick samples will be collected using a 600-micron mesh rectangular kick net. Kick samples are appropriate due to the flooding potential of the Nemadji River at these sites. The Nemadji River lacks gravel or cobble substrate at most sites. Where appropriate, woody debris will be sampled, as it is the best available alternative substrate. Exact locations for the sampling of woody debris will be made in the field at the discretion of the experienced stream biologist in order to obtain the best representation of the macroinvertebrate community present. All macroinvertebrates will be collected following *WDNR Guidelines for Collecting Macroinvertebrate Samples from Wadable Streams*. The standard field sheet contained therein will be used.

At two of the sites, duplicate kick samples will be taken in order to estimate sampling precision.

Samples will be collected between Finn Road (19.9 miles above the mouth) and the Nemadji River mouth (See Figure 1). Sample sites were chosen based on river access and may be adjusted in the field based on access and field observations. Any changes to sample site will be noted in field notes. Sample sites will be located using a map and GPS. The coordinates of the sample sites will be recorded in the field.

Macroinvertebrate samples will be prepared on site according to *WDNR Guidelines for Collecting Macroinvertebrate Samples from Wadable Streams* following collection. Samples will be preserved in wide-mouthed jars with 80-85% alcohol solution and labeled with the following information:

- Date
- Waterbody Name
- Sample ID number
- Replicate Number
- Split-sample Designation if needed
- Collectors Name

In-situ water quality data, including temperature, pH, dissolved oxygen, and conductivity will be measured and recorded at each macroinvertebrate sampling location. Water quality instruments will be subject to calibration prior to and following each field sampling event. Additionally, water quality instruments shall be properly maintained while in the field to ensure their accuracy.

Macroinvertebrate samples will be delivered directly to Dr. Kurt Schmude at University of Wisconsin-Superior by WDNR personnel. Macroinvertebrate data will be used to calculate biotic indices and other

metrics to evaluate the health of the system and assess impairments to macroinvertebrate populations in the Lower Nemadji.

Fish

Fish community surveys will be conducted in order to identify the fish assemblage present in the Lower Nemadji. Surveys will take place during a consecutive three to four-day period in July or August 2015. All sampling should occur during low-flow periods with avoidance of inclement or stormy weather.

Surveys will be conducted in a variety of physical aquatic habitat types at five, one-mile reaches along the Lower Nemadji. Locations of sample reaches will be chosen in July 2015 during a reconnaissance outing of the Lower Nemadji. Survey segments will be chosen based on accessibility and will include habitats representative of the Lower Nemadji River stratified across the range of land uses along the Lower Nemadji (urban, forest, hay/pasture). This method will ensure that the combination of reaches chosen result in a representative sample of fish assemblages in the Lower Nemadji River. A GPS unit will be used to map the survey reaches identified, and to navigate to them for the surveys.

The limited previous surveys on the Lower Nemadji noted limitations in electrofishing effectiveness due to fairly deep and turbid water; many fish escaped capture. In order to ensure that the surveys produce results representative of the entire fish community present, the surveys will be conducted with passive and active fish capture methods. Electrofishing will occur using a boat-mounted pulsed DC electrofishing boom at each survey site. The run time will be recorded for each survey. Fish immobilized during each electrofishing run will be dip-netted and put into a large tub for processing. Gill nets, minnow traps, and mini-fyke nets will also be used in each survey reach. Selection of the most appropriate gear type(s) for a particular segment will be at the discretion of the experienced on-site fisheries biologist. Major habitat types present in each reach will be targeted for sampling. Nets will be set at a rate of approximately 3 nets per mile (15 nets set total over the five sampling reaches).

Fish removed from nets will be placed in a large tub for processing. Net set time and run times shall be recorded for each net location, in order to calculate catch per unit effort. Additionally ancillary information that will be recorded for net samples will include water depth, in-stream habitat characteristics, and relative water flow characteristics.

As soon as fish are obtained via active collection methods or removed from passive collection devices, the following data will be recorded:

- Taxonomic identification to species
- Count
- Length
- Gender

Species identification will be conducted only by experienced personnel knowledgeable of the taxonomy of species in the Nemadji River. Each fish will be measured to the nearest tenth of an inch. Following processing, collected fish will be released to the source water body.

In addition to the above, the following will be recorded for each sampling event:

- Date
- Start time, Stop time
- Weather

- Field team members
- Sample collection method
- Sample Location

Data collected will be entered into the WDNR database and reviewed for errors and outliers. A data summary will be shared with the PI following completion of data collection. Fish survey data will be used to calculate established fish biotic indices to evaluate the health of the system and assess impairments to fish populations in the Lower Nemadji following data collection.

Water Chemistry

One sample will be collected each month from May to October, 2015 at three sites on the lower Nemadji River, for a total of 18 samples. Samples will be collected on the second Wednesday of each month. Sample sites will be located at State Hwy 53 (0.7 miles above mouth, SWIMS Station 163049), at Woodlawn Rd (5.4 miles above mouth, SWIMS Station 10037076), and at CTH C (11.9 miles above mouth, SWIMS Station 163003).

The following will be measured and recorded on the lab slip in the field using a water quality probe and transparency tube:

- Dissolved oxygen
- pH
- Temperature
- Conductivity
- Transparency

Table 2 shows the Lab analytical method to be used for each parameter, the container needed, preservation requirements, and maximum holding times.

Water quality instruments will be subject to calibration prior to and following each field sampling event per DNR standard operating procedures. Additionally, water quality instruments shall be properly maintained while in the field to ensure their accuracy.

Samples will also be collected for lab analysis following the WDNR Surface Water Assessment Team Guidelines and Procedures for Surface Water Grab Sampling. Samples will be taken with a Kemmerer water sampler, a discrete depth sampler. This type of sampler is lowered from the bridge in a vertical position to the desired depth, allowing water through the cylinder. A messenger is sent down to release the stoppers and close the cylinder, which is then raised. Sterile sample containers will be filled via the valve at the bottom of the Kemmerer sampler. The Wisconsin State Laboratory of Hygiene (WSLH) will provide all water chemistry sampling bottles with appropriate preservatives and shipping containers for the project.

Water samples will be collected for the following parameters:

- Total Phosphorus
- Total Kjeldahl Nitrogen
- Nitrate + Nitrite
- Ammonia
- Orthophosphate
- Total Suspended Solids

- Turbidity
- E. coli

Samples to be analyzed for TP, TKN, NO₃+NO₂, and NH₄ will be collected in one bottle and preserved with H₂SO₄. The laboratory sample bottles will be labeled with the following:

- Field identification number
- Waterbody and site name (“Nemadji R. @ CTH C”)
- Check boxes for “Filtered” and “Preserved”

Parameter	Analytical Lab Method Number	Container	Preservation Requirements	Max. Holding Times	Detection Limit
Total Phosphorus	ESS INO METHOD 310.2	250 mL	Add at least 2.0 ml H ₂ SO ₄ to pH <2 (pre-measured by WSLOH); Store on ice to be transported to the WSLOH	28 days	0.005 mg/l
Total Kjeldahl Nitrogen	ESS INO METHOD 230.3	250 mL	Add at least 2.0 ml H ₂ SO ₄ to pH <2 (pre-measured by WSLOH); Store on ice to be transported to the WSLOH	28 days	0.11 mg/l
Nitrate and Nitrite	ESS INO METHOD 220.3	250 mL	Add at least 2.0 ml H ₂ SO ₄ to pH <2 (pre-measured by WSLOH); Store on ice to be transported to the WSLOH	28 days	0.019 mg/l
Ammonia	ESS INO METHOD 220.3	250 mL	Add at least 2.0 ml H ₂ SO ₄ to pH <2 (pre-measured by WSLOH); Store on ice to be transported to the WSLOH	28 days	0.015 mg/l
Dissolved Reactive Phosphorus (Orthophosphate)	ESS INO METHOD 310.3	250 mL	Store on ice to be transported to the WSLOH	48 hours	0.00170 mg/l
Total Suspended Solids	ESS INO METHOD 340.1	1 Q	Store on ice to be transported to the WSLOH	7 days	2 mg/L
Turbidity	ESS INO METHOD 380.3	1 Q	Store on ice to be transported to the WSLOH	48 hours	0.1 NTU
E. coli	ESS MICRO METHOD 300	250 mL (2 aliquots to be collected)	Store on ice	8 hours	1 per 100 ml

Samples to be analyzed for DP will be collected in one bottle and preserved with H₂SO₄. The laboratory sample bottles will be labeled with the following:

- Field identification number

- Waterbody and site name (“Nemadji R. @ CTH C”)
- Notation that bottle will be lab-filtered.

All samples will be preserved on ice while in the field and shipped on ice to the WSLOH following WDNR and WSLOH protocols. Lab slips are linked to sample bottles with the field identification number and site name. Lab slips include:

- Date of sample (MM-DD-YYYY)
- Sample time (HH:MM, on a 24-hr clock)
- Field identification number
- Site name
- Analyses to be performed (e.g. Total Suspended Solids, etc.)
- Field parameter measurements

Two field blanks and two duplicate samples will be collected. A field blank is deionized water which is treated as a sample. It is used to identify errors or contamination in sample collection and analysis. The duplicate samples will be used to estimate sampling and laboratory analysis precision. A random number generator will be used to determine which sampling events and locations will include duplicate samples.

Grab samples will be handled and shipped to WSLOH according to the protocols specified by the WSLOH.

Water samples for E. coli analysis will be hand delivered to UWS Laboratory for analysis. Samples will be analyzed by UWS Laboratory because of its proximal location, in order to ensure that samples are analyzed within the short holding time (8 hours). An extra aliquot of water will be collected and sent to the lab. If results of the E. coli analysis for that sampling event are greater than 235 cfus, then the extra sample will be filtered and frozen by the UWS Laboratory. At the end of the season, if funds are available for analysis, a subset of the extra aliquots may be analyzed to identify the bacterial source based on DNA analysis.

Analytical and Data Analysis Requirements

Macroinvertebrates

For identification, macroinvertebrate samples will be delivered to Dr. Kurt Schmude at University of Wisconsin Superior, a regional expert on aquatic macroinvertebrates. Organisms will be identified to the lowest taxonomic level using appropriate keys (e.g. Brinkhurst 1986, Wiggins 1996, Thorp and Covich 2010), enumerated, and recorded on an identification data sheet. Data will be delivered to the PI and reviewed for outliers or irregular results. Any questionable data will be flagged and requested further review by the Schmude Lab as necessary. Data will be entered into DNR’s SWIMS database. The macroinvertebrate calculator in SWIMS will generate the Macroinvertebrate Index of Biological Integrity (MIBI), the Hilsenhoff Biotic Index (HBI), and various other macroinvertebrate metrics using the standard methods described in Weigel (2003) and Lillie et al. (2003). The biotic indices will be used to evaluate the health of the macroinvertebrate community in the Lower Nemadji.

Fish

Data collected will be entered into the WDNR database and reviewed for errors and outliers. A data summary will be shared with the PI following completion of data collection. Fish survey data will be used to calculate established fish biotic indices using methods developed by Lyons et. al. (2001) for Wisconsin’s large warmwater rivers. The IBI was developed through a standardized electrofishing

method, and the number of native species, number of sucker species, number of intolerant species, percent composition of lithophils, and percent composition of insectivores were among the metrics used to calculate IBI scores. Due to the potential for reduced electrofishing capture efficiency in the Nemadji River, the IBI will be calculated from multiple fish capture gear standardized at three individual survey locations in each of five one-mile river segments. This standardized design is anticipated to offset any variability from using multiple fish survey gear. The IBI score will correspond with qualitative fish community ratings such as good, fair, poor that will be used to describe the overall condition of the Nemadji River fish community.

Water Quality

Samples will be sent to WSLOH for analysis. Table 2 shows the Lab analytical method to be used for each parameter, the container needed, the preservation requirements, and the maximum holding times. Samples run after the holding time will be qualified in results. Data collected will be transmitted electronically into the SWIMS database by WSLOH once results are available. The PI will review the data and check for outliers. Ranges and mean concentrations for each parameter for each site and for the entire Lower Nemadji will be calculated. The data and data limitations will be summarized as part of the final report. If macroinvertebrate and/or fish communities are evaluated to be under-performing, or are not as healthy as expected, then results of the biotic surveys will be compared with water quality results to determine if there are any significant correlations.

Quality Control Requirements

In general, quality control measures for all types of sampling for this project will include following standard DNR protocol during sampling, choosing suitable analytical procedures, and following all standard laboratory protocols and QC procedures. This will allow comparison of this dataset with other datasets with consistent documented sampling methods and conditions and analytical procedures.

Because of the difference in data collection methods as well as significant difference in ecological conditions present in the Lower Nemadji compared to the lower St. Louis River estuary, the data collected through this effort will *not* be included in the St. Louis River Area of Concern Benthic Macroinvertebrate Index.

Completeness of the resultant datasets will be measured by dividing the number of valid (passing QA/QC requirements) measurements actually obtained by the number of measurements made. The completeness requirement has been set at 90 %.

Macroinvertebrate

Sample locations will be chosen to be representative of all the habitats present in the Lower Nemadji River. Macroinvertebrate sampling will be conducted in the fall, because the populations sampled may be less influenced by short-term spring flood events and more representative of the river. Additionally, sites are easier to access due to lower water levels.

Appropriate sampling methods will be used for a riverine setting (kick nets and woody debris samples). Two duplicate kick samples will be collected for macroinvertebrates. During water quality sampling, two duplicate samples will be collected, which represents 10% of samples collected. The duplicate sample will be used to estimate sampling and laboratory analysis precision. Table 2 below shows the control limits that will be applied to the analyses. WDNR standard sampling protocols will be used, along with standard preservation, and shipping methods. Sample analysis will be conducted by the Dr. Kurt Schmude at UWS, a state-wide expert in aquatic invertebrate taxonomy. Standard protocol will be used

to identify macroinvertebrates down to the lowest possible taxonomic level. Data will be reviewed according to standard lab protocol before delivery to WDNR. Data will then be reviewed for outliers or other irregular data by WDNR staff.

Fish

Sampling sites for fish surveys will be chosen in June, 2015 based on site visits by the experienced staff fish biologist. Survey reaches will include all habitat types in the Lower Nemadji, stratified across the range of land use types in the lower Nemadji watershed. The land use (forest, urban, agriculture) could influence the quality of in-stream habitat, so to get a representative sample of fish in the river, sample sites will be stratified across all types of land use. A range of capture techniques will be used in order to ensure that a representative population of fish is sampled. Data will be reviewed for outliers or other irregular data by WDNR staff.

Water Quality

Water quality samples aim to represent average water quality of the main stem of the Lower Nemadji River. Sample sites were chosen across the study area based on access. Studies have shown that using a modified-random sampling design based on stream-crossing road access sites results in no statistically significant difference in results compared to a true random sampling design (Miller et al, 2009). The water quality samples will be taken from the middle of the river using a Kemmerer sampler, in order to ensure well-mixed water that is representative of the lower main stem of the river. Samples will be taken below the surface to avoid influences from surface water.

Two field duplicates (10% of all samples) will be collected can be used to ascertain the variance of the data due to field and laboratory protocols. Additionally, two field blanks of deionized water will be collected and treated as a sample. The field blank is used to identify errors or contamination in sample collection and analysis.

WDNR standard sampling protocols will be used, along with standard preservation, and shipping methods will be used. DNR staff will ensure that preservation requirements are met and holding times are not exceeded in the laboratory. The WSLOH has standard procedures for instrument calibration quality control, which are found in the *Quality Assurance Manual - Environmental Health Division - Wisconsin State Laboratory of Hygiene*. Table 2 below shows the control limits that will be applied to the analyses.

Data will be reviewed by the State Lab of Hygiene as per WSLOH standard QC protocol. Data will be again reviewed for outliers or other irregular data by WDNR staff.

C. Assessment/Oversight and Data Validation

Reports to Management

The project coordinator is responsible for quarterly reporting for this grant to the EPA. Quarterly reports will be entered into the SWIMS project by the 15th of the month following the end of the quarter.

A final report will be produced by the PI with assistance from the fish biologist summarizing the results of this monitoring. The final report will include the following:

- Results of Macroinvertebrate IBIs and interpretation of health of macroinvertebrate community on the lower Nemadji River

- Results of the Fish IBIs and interpretation of health of fish community on the Lower Nemadji River
- Potential water quality or other environmental stressors related to any observed impairments in macroinvertebrate and fish community health.

This report will be used to inform and direct future management actions related to the Excessive Loading of Sediment and Nutrients beneficial use impairment on the Nemadji River in the St. Louis River Area of Concern.

Data Review, Validation, or Verification

Macroinvertebrates and Fish

During fish and macroinvertebrate sampling, field staff will assess efforts to ensure data is collected as described in the Quality Assurance Project Plan. If macroinvertebrates or fish are not able to be collected using the methods outlined here, field methods will be modified. The PI and project biologists will use their expert discretion to adapt field methods as necessary to complete surveys. Any changes to the methods outlined here will be documented in field notes and included in the final report.

After macroinvertebrate and fish data is entered into DNR databases, the PI will review data to determine whether there are potential problems with the data. If there are problems with macroinvertebrate or fish data, corrective action will be taken. DNR may request the Schmude lab to review samples and re-analyze samples if necessary. If problems are identified with the fish data, re-sampling will be considered and completed as needed.

Water Quality

During the field season, timing of the sampling will be reviewed periodically to see if the regular sampling on the second Wednesday of each month is resulting in a sample set that is representative of the average water quality in the Nemadji River. If necessary (for example, if sampling events are biased towards high-flow events), the sampling time will be adjusted to ensure a representative dataset.

The field blanks should be at or below the laboratory detection limits. If not, equipment used to retrieve, store and process samples will be cleaned and checked to determine the cause, and appropriate corrective actions will be taken; plus, another field blank will be collected and analyzed to confirm that corrective actions were sufficient.

The laboratory is assessed as part of the routine laboratory certification process. The laboratory will review their data as part of their normal verification process prior to issuing the report. When results are received from the laboratory, they will be reviewed for any obvious errors or results that may be outliers. Depending on the situation, the laboratory may be requested to reanalyze the sample or examine the sample for any attributable cause. As part of the statistical assessment of the data, the data set will be checked for outliers.

Reconciliation with Data Quality Objectives

The Data quality objectives were as follows:

- DQO 1 Evaluate the health of the biological communities in the Lower Nemadji River through the use of indices of biological integrity (IBIs).
- DQO 2 If impairments are observed, connect any observed impairments in biological community health to observed and documented water quality stressors.

Macroinvertebrate surveys will be conducted at a rate of approximately one sample per two miles of river along the Lower Nemadji River in order to calculate standard biotic indices for the macroinvertebrate community based on Weigel (2003) and Lillie et al. (2003). Electrofishing as well as fish net surveys (e.g. gill net, minnow traps, fyke nets) will be conducted along five one-mile reaches of the Lower Nemadji in order to calculate biotic indices for the fish community based on Lyons et al. (2001). In order to connect any possible fish and macroinvertebrate community health impairments to water quality stressors, water quality data will also be collected along the lower reaches of the Nemadji River.

The main data user for the data generated will be water resource managers within the DNR. The biotic indices calculated from fish and macroinvertebrate data will result in estimates of community health from “poor” to “excellent” at each survey site. As a whole, these will allow resource managers to determine if the fish and macroinvertebrate communities in the Lower Nemadji are healthy or impaired. If resource managers determine that the communities are under-performing, or are not as healthy as expected, then results of fish and macroinvertebrate surveys will be compared with water quality results to determine if there are any significant correlations. The results of this assessment will provide resource managers in the St. Louis River Area of Concern with information needed to understand if the biological community in the Lower Nemadji River is poor, fair, good, or excellent and if there are potential connections between water quality and any observed impairments. This assessment, along with the results of a Nemadji River watershed sediment loading study and a Nemadji River watershed Hydrological Simulation Program--Fortran (HSPF) modelling study will allow resource managers to better understand the sediment- and nutrient-loading impairments in the Nemadji River watershed and determine if additional management actions are needed in order to remove the Excessive Sediment and Nutrient Loading beneficial use impairment in the St. Louis River Area of Concern.

D. References

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