2016 Reference Site Monitoring Work Plan

PREPARED BY

Kevin Gaston
Probabilistic Monitoring Section

WATERSHED ASSESSMENT and PLANNING BRANCH (WAPB)
Indiana Department of Environmental Management (IDEM)
Office of Water Quality (OWQ)
100 North Senate Avenue
MC65-40-2 Shadeland
Indianapolis, Indiana 46204-2251

December 28, 2016

B-030-OWQ-WAP-PRB-16-W-R0

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SIGNATURE PAGE

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Indiana Department of Environmental Management
Office of Water Quality
Watershed Assessment and Planning Branch
Indianapolis, Indiana
B-030-OWQ-WAP-PRB-16-W-R0

Author(s) Signatures

Kevin Gaston _____ Date 12-28-16

Management Reviews and Approvals

_____ Date 1-3-2017
Stacey Sobat, Section Chief Probabilistic Monitoring Section
IDEM Watershed Assessment and Planning Branch

_____ Date 12/29/16
Cyndi Wagner, Section Chief Targeted Monitoring Section
IDEM Watershed Assessment and Planning Branch

_____ Date 1-3-2017
Timothy Bowren, Project Quality Assurance Officer, Technical and Logistical Services Section
IDEM Watershed Assessment and Planning Branch

_____ Date 12/29/16
Mike Sutton, Section Chief Technical and Logistical Services Section, Quality Assurance Manager
IDEM Watershed Assessment and Planning Branch

_____ Date 12/28/16
Marylou Renshaw, Branch Chief, Branch Quality Assurance Coordinator
IDEM Watershed Assessment and Planning Branch

IDEM Quality Assurance Staff reviewed and approves this Sampling and Analysis Work Plan.

_____ Date 1/3/17
Quality Assurance Staff
IDEM Office of Program Support

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WORK PLAN ORGANIZATION

This Sampling and Analysis Work Plan is an extension of the existing Watershed Assessment and Planning Branch, October 2004 "*Quality Assurance Project Plan (QAPP) for Indiana Surface Water Quality Monitoring and Total Maximum Daily Load (TMDL) Program*" and serves as a link to the existing QAPP as well as an independent QAPP of the project. As per the United States Environmental Protection Agency (U.S. EPA) 2006 QAPP guidance (U.S. EPA 2006), this Work Plan establishes criteria and specifications pertaining to a specific water quality monitoring project that are usually described in the following four groups (phases) or sections as QAPP elements:

Section I. Project Management/Planning

- Project Objective
- Project/Task Organization and Schedule
- Background and Project/Task Description
- Data Quality Objectives (DQOs)
- Training and Staffing Requirements

Section II. Measurement/Data Acquisition

- Sampling Procedures
- Analytical Methods
- Sample and Data Acquisition Requirements
- Quality Control (QC) Measures Specific to the Project

Section III. Assessment/Oversight

- External and Internal Checks
- Audits
- Data Quality Assessments (DQAs)
- Quality Assurance/Quality Control (QA/QC) Review Reports

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- Data Handling and associated QA/QC activities
- QA/QC Review Reports

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LIST OF ACRONYMS

AIMS:	Assessment Information Management System
ASTM:	American Society for Testing and Materials
AU:	Assessment Unit
BSS:	Biological Studies Section
CAC:	Chronic Aquatic Criterion
CALM:	Consolidated Assessment Listing Methodology
CCC:	Criterion Continuous Concentration
CFR:	Code of Federal Regulations
CPR:	Cardio-Pulmonary Resuscitation
DELT:	Deformity, Eroded Fin, Lesion, Tumor
DIC:	Differential Interference Contrast (Microscope)
DQA:	Data Quality Assessment
DQO:	Data Quality Objective
GPP:	Generator Powered Pulsator
GPS:	Global Positioning System
HDPE:	High-density polyethylene
IAC:	Indiana Administrative Code
IBI:	Index of Biotic Integrity
IC:	Indiana Code
IDEM:	Indiana Department of Environmental Management
MHAB:	Multi-habitat
MIBI:	Macroinvertebrate Index of Biotic Integrity
MS/MSD:	Matrix Spike/Matrix Spike Duplicate
NHD:	National Hydrography Database
NHEERL:	National Health Environmental Effects Research Laboratory
NTU:	Nephelometric Turbidity Unit(s)
OHEPA:	Ohio Environmental Protection Agency
OWQ:	Office of Water Quality
PFD:	Personal Floatation Device
PPE:	Personal Protective Equipment
QA:	Quality Assurance
QA/QC:	Quality Assurance/Quality Control
QC:	Quality Control
QAPP:	Quality Assurance Project Plan
QHEI:	Qualitative Habitat Evaluation Index
RPD:	Relative Percent Difference
SM:	Standard Methods
SOLAS:	Safety of Life at Sea
SOP:	Standard Operating Procedures
S.U.:	Standard Units
TMDL:	Total Maximum Daily Load
U.S. EPA:	United States Environmental Protection Agency
USGS:	Unites States Geological Survey
WAPB:	Watershed Assessment and Planning Branch

DEFINITIONS

Elutriate	To purify, separate, or remove lighter or finer particles by washing, decanting, and settling.
Fifteen (15) Minute Pick	A component of the IDEM multihabitat macroinvertebrate sampling method in which the one minute kick sample and fifty meter sweep sample collected at a site are combined, elutriated, with macroinvertebrates removed from the resulting sample for 15 minutes while in the field.
Fifty (50) Meter Sweep	A component of the IDEM multihabitat macroinvertebrate sampling method in which approximately 50 meters (50m) of shoreline habitat in a stream or river is sampled with a standard 500 micrometer (500 µm) mesh width D-frame dipnet by taking 20-25 individual “jab” or “sweep” samples, which are then composited.
Macroinvertebrate	Aquatic animals which lack a backbone, are visible without a microscope, and spend some period of their lives in or around water.
One (1) minute kick sample	A component of the IDEM multihabitat macroinvertebrate sampling method in which approximately one square meter (1 m^2) of riffle or run substrate habitat in a stream or river is sampled with a standard 500 micrometer (500 µm) mesh width D-frame dipnet for approximately one (1) minute.
Ocular Reticle	A thin piece of glass marked with a linear or areal scale that is inserted into a microscope ocular, superimposing the scale onto the image viewed through the microscope.
Perihyton	Algae attached to an aquatic substrate.
Reach	A segment of a stream used for fish community sampling equal in length to 15 times the average wetted width of the stream, with a minimum length of 50 meters and a maximum length 500 meters.
Seston	Organic matter suspended in the water column generally comprised of phytoplankton, bacteria and fine detritus.

I. PROJECT MANAGEMENT/PLANNING

Project Objective

The objective of the 2016 Reference Site Monitoring Project is to provide physical, chemical, and biological data from reference sites that will be used to develop/refine the Index of Biotic Integrity (IBI) for aquatic assemblages (including diatoms, macroinvertebrates, and fish) as well as biological criteria for aquatic life use assessments. The IBI is composed of 12 biological assemblage characteristics or metrics that assess the aquatic communities' structural, compositional, and functional integrity. Different IBI metrics may be used depending on variables such as what part of the state you are sampling (ecoregion) and size of stream (drainage area). The 12 different metrics can score 0, 1, 3, or 5, which represents the deviation from expected community structure (i.e. 5 = no deviation from expectations, 0 = severe deviation from expected community structure). The total IBI score can range from 0 (severe disturbance) to 60 (excellent, comparable to "least impacted" conditions). For more information on the fish and macroinvertebrate IBI, view Appendix 2 and 3 at the end of this work plan.

Reference sites are located in areas with the least amount of anthropogenic disturbance and are considered the most natural remaining areas within a specified geographic boundary. Candidate sites are chosen based on abiotic factors such as land use, water chemistry, and in-stream physical habitat that function as potential stressors to the biotic components (i.e. fish/macroinvertebrate/diatom communities) of the stream or river ecosystem.

The primary filter used in selecting reference sites is land use criteria such as:

- percent of agricultural or urban areas
- impervious surface area
- human population density and distribution
- road density and crossings
- proportion of active mining activities
- proportion of protected lands
- proximity to permitted facilities, confined feeding operations, and Superfund sites.

In altered watersheds, chemical and in-stream physical habitat data may be used as a secondary filter to select reference sites and develop biological expectations for "least disturbed condition" (best available condition given widespread disturbance) rather than "minimally disturbed condition" (nearly absent human disturbance) or "historical condition" (prior to major industrialization, urbanization, and intense agricultural practices) (Stoddard et al. 2006).

Ideally, reference sites should be sampled at least once every 10 years to monitor for changes in the biological expectations for "least disturbed condition" and possible revisions to biological criteria. Sampling at reference sites should include a minimum of two biological communities (fish, macroinvertebrates, and/or diatoms), habitat evaluations, and at least *in-situ* water chemistry. Ideally, laboratory water chemistry parameters, algal biomass, and flow will be sampled additionally as resources allow.

The Indiana Department of Environmental Management (IDEM) Office of Water Quality (OWQ) worked with U.S. EPA and Tetra Tech in March 2015 to develop a framework and criteria for reference site selection (U.S. EPA Assistance Agreement I 96555711-1 IDEM). IDEM provided Tetra Tech with 1458 sites previously sampled for fish and/or macroinvertebrates between 2003 and 2013 for possible reference site selection. Using land use factors as the primary filter, Tetra Tech provided a list of 324 reference sites. IDEM narrowed down the list further by using in-stream chemical and physical data as a secondary filter. A minimum of 20 reference sites are required in each of the natural environmental gradient classifications (i.e. ecoregion, stream size, etc.) to develop linear regression models showing change in biological assemblage structure

given certain explanatory variables. Increasing the number of reference sites, however, will reduce variability in calibrating the IBI and setting biological criteria thresholds (U.S. EPA 2013, Tetra Tech personal communication). Based on the spatial distribution of the sites and available resources, IDEM will conduct site reconnaissance and sampling of reference sites with the goal of at least 20 reference sites each year over the next 10 years to refine biological indices and water quality criteria, as well as possibly develop other assessment indicators and thresholds.

Project/Task Organization and Schedule

Sampling for this project will begin in April and continue through November 2016. Chemical, physical, and biological parameters will be collected. Laboratory processing and data analysis for the project will continue through spring of 2017.

Timeframes for sampling activities include:

Activity	Date(s)	Number of Sites	Frequency of Sampling-related activity	Parameters to be sampled	How evaluated
Site reconnaissance	February through end of April	25 to ensure a minimum of 20 sites sampled during all three sampling events	Until 25 accessible target sites confirmed or April deadline reached	Safety to access stream and proper equipment for sampling	Land owner approval and best professional judgment
Biological sampling	June 6 through November 13	Minimum of 20 sites, 25 if water present in at least half the sampling reach	Once each for: Fish community (June 6-October 14), Macroinvertebrate community (July 11 – November 11) (both may occur on same day from July 11 – October 14)	Fish: Identification to species, number of individuals, minimum and maximum length, batch weight, deformities, eroded fins, lesions, tumors. Macroinvertebrates: 15 minute pick of individual specimens for diversity (later identified to lowest taxonomic level in lab) Separate Qualitative Habitat Evaluation Index (QHEI) for each community sampled	Fish and Macroinvertebrates are evaluated using the appropriate IBI. If If IBI less than 36, the site is impaired for aquatic life use. QHEI evaluated separately for fish and macroinvertebrate communities, as reaches may be of different lengths, or sampled on different days. A QHEI score of less than 51 (range 0-100) indicates that habitat may be impacting the integrity of the biological community.

Activity	Date(s)	Number of Sites	Frequency of Sampling-related activity	Parameter to be sampled	How evaluated
Water chemistry	April, June, August	25 to ensure a minimum of 20 sites sampled during all three sampling events	Three times: Once each in April, June, and August, with a minimum of 30 days between sampling events	Phosphorous Nitrogen (NO_3 & NO_2) Dissolved O_2 pH Algal conditions **From Table 1 (details below)** - Metals, dissolved - Arsenic - Un-ionized ammonia as N - Chloride - Sulfate - Dissolved Solids	**Nutrient Benchmarks:** Once@ >0.3 mg/L (for nutrients) Once@ >10.0 mg/L (for nutrients) <4.0 mg/L; >12 mg/L (for nutrients) >9.0 Standard Units (for nutrients) Excessive (for nutrients) **Water Quality Standard Limits:** 190 ug/L 750 mg/L
Algal samples	August	Minimum of 20 sites, 25 if water present in at least half the sampling reach	Once, with the 3rd water chemistry sample, August	Periphyton (Seston included if drainage area >1000 square miles) Algal diatoms	Diatom identification and enumeration Chlorophyll *a* and Pheophytin *a*

Background and Project/Task Description

The Reference Site Monitoring Project is operated through the Watershed Assessment and Planning Branch (WAPB), OWQ, IDEM. Other organizations which help with data preparation, collection, and analysis include private laboratories under contract with the State of Indiana (Request For Proposals 12-48, see IDEM 2012a), United States Geological Survey (USGS), Department of Biological and Environmental Sciences at Georgia College and State University, U.S. EPA Region V, and the Indiana Department of Natural Resources. Landowners and property managers throughout the State also participate in the Reference Site Monitoring Project by assisting staff in accessing remote stream locations to collect various sampling media.

The Reference Site Monitoring Project provides physical, chemical, and biological data that will be used to calibrate the IBI for aquatic assemblages. This is accomplished by sampling reference sites in Indiana over 10 years to assess and characterize overall water quality and biological integrity; thus, the project will be continuous as refinements to the IBI calibration will be documented every 10 years. Data from the following parameters will be investigated and utilized for IBI and biological criteria refinement as well as assessment purposes: water chemistry and

algal samples (seston and periphyton), fish/macroinvertebrate/diatom assemblages, and habitat evaluations.

Data Quality Objectives (DQO)

The DQO process (U.S. EPA 2006) is a planning tool for data collection activities. It provides a basis for balancing decision uncertainty with available resources. The DQO is required for all significant data collection efforts for a project and is a seven-step systematic planning process used to clarify study objectives, define the types of data needed to achieve the objectives, and establish decision criteria for evaluating data quality. The DQO process for the 2016 Reference Site Monitoring Project is identified in the following seven steps:

1. State the Problem

Indiana is required to assess all waters of the State to determine their designated use attainment status**. "**Surface waters of the State are designated for full-body contact recreation" and "will be capable of supporting" a "well-balanced, warm water aquatic community" [327 Indiana Administrative Code (IAC) 2-1-3 (for waters outside the Great Lakes system)] and [327 IAC 2-1.5-5 (for waters within the Great Lakes system)]. This project will gather biological (algal, fish and macroinvertebrate), chemical, and habitat data at reference sites for the purpose of refining the IBI metrics and biological criteria thresholds in the State of Indiana to more accurately assess aquatic life use attainment status.

2. Identify the Decision

The objective of this project is to sample reference sites throughout Indiana to provide biological assemblage information with chemical and physical parameters that will be used to refine or validate IBI metrics and biological criteria thresholds every 10 years.

Chemical and physical sampling data should be used to validate the absence of anthropogenic disturbance or a minimal level of allowed disturbance at reference sites (U.S. EPA 2013); thus, each site will be evaluated as "supporting" or "non-supporting" when compared with water quality criteria shown in Table 1 [Non-Great Lakes, 327 IAC 2-1-6] and [Great Lakes, 327 IAC 2-1.5-8] following Indiana's 2014 Consolidated Assessment Listing Methodology (CALM, IDEM 2014a pages 24-28).

In addition to the physical and chemical criteria listed in Table 1, data for several nutrient parameters will be evaluated with the benchmarks listed below (IDEM 2014a). Assuming a minimum of three sampling events, if two or more of the conditions below are met on the same date, the assessment unit of the waterbody will be classified as non-supporting due to nutrients.

- Total Phosphorus: one or more measurements >0.3 mg/L
- Nitrogen (measured as NO_3+NO_2): one or more measurements >10.0 mg/L
- Dissolved Oxygen (DO):
    - For cold-water fish, <6.0 mg/L or <7.0 mg/L in areas where spawning occurs during the spawning season and in areas used for imprinting during the time salmonids are being imprinted.
    - For warm water, <4.0 mg/L or measurements consistently at or close to the standard, ranging from 4.0-5.0 mg/L or >12.0 mg/L
- pH: >9.0 Standard Units (S.U.) or measurements consistently at or close to the standard, ranging from 8.7-9.0 S.U.
- Algal Conditions: visually observed as "Excessive" by trained staff using best professional judgment

<u>Biological Criteria:</u>
Indiana narrative biological criteria [327 IAC 2-1-3] and [327 2-1.5-5] states that "all waters, except as described in subdivision (5)," (i.e., limited use waters) "will be capable of supporting" a

"well-balanced, warm water aquatic community". The water quality standard definition of a "well-balanced aquatic community" is "an aquatic community that: (A) is diverse in species composition; (B) contains several different trophic levels; and (C) is not composed mainly of pollution tolerant species" [327 IAC 2-1-9] and [327 IAC 2-1.5-2]. An interpretation or translation of narrative biological criteria into numeric criteria would be as follows: A stream segment is non-supporting for aquatic life use when the monitored fish or macroinvertebrate community receives an IBI score of less than 36 which is considered "Poor" or "Very Poor" (IDEM 2014a). IBI scores greater than or equal to 36 are considered "Fair" to "Excellent" on the scale of 0-60; thus, the stream segment is supporting aquatic life use.

Assessment of each site sampled will be reported to U.S. EPA in the 2018 update of Indiana's Integrated Water Monitoring and Assessment Report. Site specific data will be used to classify associated assessment units (AU) into one of five major categories in the State's Consolidated list, category definitions are available in the Integrated Report (IDEM 2014b, p. 64).

Sites evaluated as non-supporting for aquatic life use will be rejected as a reference site, initially decreasing the number of reference sites in the State of Indiana. After 10 years, however, IDEM may discover additional reference sites by reviewing land use criteria as well as chemical and in-stream physical habitat data for sites with biological assemblage information sampled through additional projects between 2014 and 2024.

To develop the IBI for diatoms as well as biological criteria for aquatic life use assessments, benthic diatoms will be collected in conjunction with chemical and chlorophyll *a* data from each site along with field parameters and physical site descriptions. Once collected, the samples will be preserved and transported to the laboratory where algae will be identified and enumerated as part of the development of algal metrics.

Table 1. Water Quality Criteria [Non-Great Lakes, 327 IAC 2-1-6] and [Great Lakes, 327 IAC 2-1.5-8].

Parameter	Level	Criterion
Metals (dissolved)	Calculated based on hardness	CAC (Non-Great Lakes) CCC (Great Lakes)
Arsenic III (dissolved)	190 µg/L 147.9 µg/L	CAC (Non-Great Lakes) CCC (Great Lakes)
Ammonia as Nitrogen	Calculated based on pH and temperature	CAC (Non-Great Lakes) CCC (Great Lakes)
Chloride	Calculated based on hardness and sulfate	CAC (Non-Great Lakes) CCC (Great Lakes)
Dissolved Oxygen (DO)	At least 5.0 mg/L (warm water aquatic life) At least 6.0 mg/L (cold-water fish*)	Not less than 4.0 mg/L at any time. Not less than 6.0 mg/L at any time and shall not be less than 7.0 mg/L in areas where spawning occurs during the spawning season and in areas used for imprinting during the time salmonids are being imprinted.
pH	6.0 - 9.0 S.U.	Must remain between 6.0 and 9.0 S.U. except for daily fluctuations that exceed 9.0 due to photosynthetic activity
Nitrate-N+Nitrite-N	10 mg/L	Human Health point of drinking water intake

Sulfate	Calculated based on hardness and chloride (Non-Great Lakes) 250 mg/L (Great Lakes)	In all waters outside the mixing zone Public water supply
Dissolved Solids	750 mg/L	Public water supply

CAC = Chronic Aquatic Criterion, CCC = Criterion Continuous Concentration, S.U. = Standard Units

*Waters protected for cold-water fish include those waters designated by the Indiana Department of Natural Resources for put-and-take trout fishing as well as salmonid waters listed in 327 IAC 2-1.5-5.

3. Identify the Inputs to the Decision

Field monitoring activities are required to collect physical, chemical, algal, biological and habitat data. Samples will be collected for physical and chemical parameters as well as algal and biological communities if the flow is not dangerous for staff to enter the stream and barring any hazardous weather conditions or unexpected physical barriers to access the site. Even if the weather conditions and stream flow are safe, sample collections for algal and biological communities may also be postponed at a particular site for one to four weeks due to scouring of the stream substrate or instream cover following a high water event resulting in non-representative samples. Collection procedures for field measurements, algal, chemical, biological, and habitat data will be described in detail under Section II MEASUREMENT/DATA ACQUISITION.

4. Define the Boundaries for the Study

In 2016 reference sites will be sampled in two separate areas of the state to allow flexibility in sampling as a result of high water or bad weather preventing sampling in IDEM projects in other areas of the state. Further explanation of site selection is explained in Sampling Design and Site Locations in Section II. Measurement/Data Acquisition of this work plan. Table 2 and Figure 1 provide location information for reference sites sampled in 2016 from the Northern Basin. Table 3 and Figure 2 provide location information for reference sites sampled in 2016 from the Southern Basin.

5. Develop a Decision Rule

All sites will be evaluated for aquatic life use support (ALUS) decisions. For assessment purposes in the Indiana Integrated Water Monitoring and Assessment Report, aquatic life use support decisions will include independent evaluations of chemical and biological criteria as outlined in Indiana's 2014 CALM (IDEM 2014a, pages 24-28). The fish and macroinvertebrate assemblage will be evaluated at each site using the appropriate IBI. Specifically, a site will be considered non-supporting for aquatic life use when IBI scores are less than 36. Given more recent data, assessment decisions will be reported in the 2018 Indiana Integrated Water Monitoring and Assessment Report resulting in stream segments being de-listed for impaired biotic community (now fully supporting aquatic life use) or listed as non-supporting for aquatic life use due to a change in water quality or habitat that has impaired the biotic community.

Sites not supporting aquatic life use or sites violating the minimal allowable amount of disturbance will be rejected as reference sites due to chemical or physical alterations detected by

current sampling efforts. To avoid circularity in deriving IBI calibrations, reference sites will not be chosen based on biological attributes (i.e. excellent IBI metrics or total scores).

IDEM's intention is to use algal metrics, once determined, as part of nutrient criteria being developed for Indiana's surface waters. Eventually, IDEM also plans to use algal metrics with macroinvertebrate and fish metrics for ALUS decisions. Given that ecological tolerances for many diatom species are known, changes in diatom community composition can be used to diagnose the environmental stressors affecting ecological health (Stevenson 1998; Stevenson and Pan 1999); thus, periphyton IBI metrics have been developed and tested in many regions (Kentucky Department of Environmental Protection 1993; Hill 1997). The periphyton assemblage may be used to assess biological integrity of a waterbody without any other information; however, periphyton are most effective when used with habitat and macroinvertebrate assessments particularly because of the close relationship between periphyton and these elements of stream ecosystems (Barbour et al. 1999). For this reason, algal sampling will be conducted at the same sites where macroinvertebrates, fish, habitat, chemical, and physical data will be collected as part of the Reference Site Monitoring Program.

Table 2. List of 2016 Reference Sites for the Northern Basin.

Site #	AIMS Site Name	Event ID	Stream Name and Location	County	Hydrologic Unit Code (HUC)	Topo	Latitude (DD)	Longitude (DD)	Stream Order	Drainage Area (mi^2)	Gradient (ft/mi)
28	LMG-04-0039	16R028	East Branch Little Calumet River @ Ottis Road	LaPorte	040400010401	A-34	41.612775	-86.906092	2	8.202	20.54
29	LMG070-0035	16R029	East Branch of Trail Creek @ CR 700 N	LaPorte	040400010104	A-12	41.706571	-86.770572	2	5.7	14.67
31	LMG100-0009	16R031	Tributary of Spring Creek @ CR 1000 N	LaPorte	040400010204	A-01c	41.752902	-86.560427	2	3.77	19.96
32	UMK060-0041	16R032	Eagle Creek @ CR 700 E	Starke	071200010504	B-09	41.303940	-86.563448	3	31.36	14.5
33	UMK060-0042	16R033	Yellow River @ 13 B West	Marshall	071200010505	B-10	41.276518	-86.449617	3	368	3.1
34	LMJ230-0008	16R034	Baugo Creek @ Ash Road	Elkhart	040500012104	A-18	41.648926	-86.055623	4	76.23	7.58
35	LMJ190-0031	16R035	Elkhart River @ CR 127	Elkhart	040500011805	A-43	41.500997	-85.779920	3	377.13	5.77
36	LMJ120-0042	16R036	Pigeon River @ N 675 W	LaGrange	040500011106	A-22	41.740613	-85.557945	4	335.5	3.2
37	LEJ080-0014	16R037	Tributary of Leins Ditch @ CR 12	DeKalb	041000030601	A-71	41.480792	-85.137590	1	27.79	41.48
38	LEJ050-0011	16R038	Fish Creek @ CR 18	DeKalb	041000030406	A-74	41.463611	-84.811389	4	3.939	41.46
39	LEJ090-0041	16R039	Black Creek @ CR 9	DeKalb	041000030704	B-20	41.282420	-85.142408	2	5.878	41.28

Table 3. List of 2016 Reference Sites for the Southern Basin.

Site #	AIMS Site Name	Event ID	Stream Name and Location	County	Hydrologic Unit Code (HUC)	Topo	Latitude (DD)	Longitude (DD)	Stream Order	Drainage Area (mi^2)	Gradient (ft/mi)
40	WEM080-0023	16R040	Mutton Creek @ CR 600 W	Jennings	051202070704	H-17	39.042128	-85.722319	1	2.6	21.21
41	WEM020-0015	16R041	Graham Creek @ CR 500 S	Jennings	051202070204	H-41	38.916293	-85.598847	3	84.41	4.92
42	OSK060-0001	16R042	Bull Creek @ Blue Ridge Road	Clark	051401010603	I-59	38.481086	-85.513601	1	3.8	61.00
43	WEM060-0044	16R043	Otter Creek @ CR 500 E	Jennings	051202070303	H-18	39.003397	-85.520050	2	62.88	10.34
44	WEM050-0048	16R044	Vernon Fork Muscatatuck River @ CR 740 E	Jennings	051202070402	G-65	39.136189	-85.475837	2	41.82	11.98
45	WEM050-0040	16R045	Finch Branch @ CR 830 E	Jennings	051202070403	H-19	39.097528	-85.470622	1	2.92	18.31
46	WEM020-0030	16R046	Rush Branch @ W Perimeter Road	Jennings	051202070203	H-42	38.993017	-85.467331	1	2.51	27.71
47	WEM020-0040	16R047	Little Graham Creek @ W Perimeter Road Area 33	Jennings	051202070202	H-42	38.938317	-85.462419	2	20.91	16.71
48	OSK030-0019	16R048	West Fork Indian Kentuck Creek @ N China Manville Road	Jefferson	051401010204	H-66	38.809436	-85.316409	2	36.5	15.90
49	OSK030-0017	16R049	Indian Kentuck Creek @ Lonnis Hill Road	Jefferson	051401010205	H-66	38.844077	-85.25926279	3	50.7	18.30
50	OSK-02-0016	16R050	Indian Kentuck Creek @ Brooksburg Manville Road	Jefferson	051401010206	H-67	38.758409	-85.247814	3	128.842	3.41
51	OSK-02-0017	16R051	Lost Fork Creek @ Lost Fork Road	Jefferson	051401010206	I-20	38.721648	-85.207689	1	5.179	33.33
52	WEM050-0034	16R052	Vernon Fork Muscatatuck River @ CR 1220 N	Jennings	051202070402	G-65	39.163292	-85.462400	2	36.98	8.72
53	OSK-02-0023	16R053	West Fork Indian Kentuck Creek @ Wolf Run Road	Jefferson	051401010204	H-66	38.789341	-85.284761	2	46.01	19.05

Figure 1. 2016 Reference Sites for the Northern Basin.

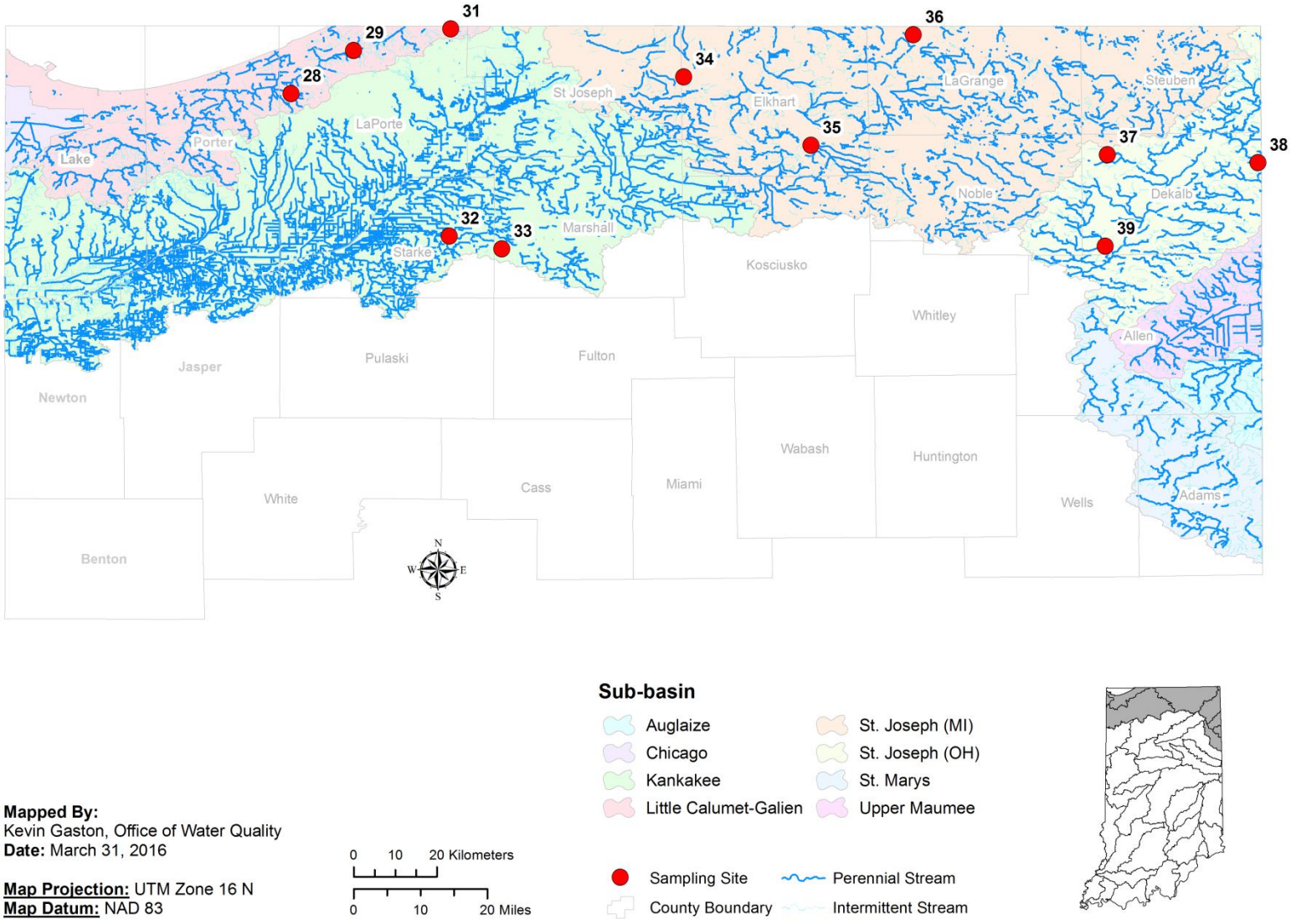
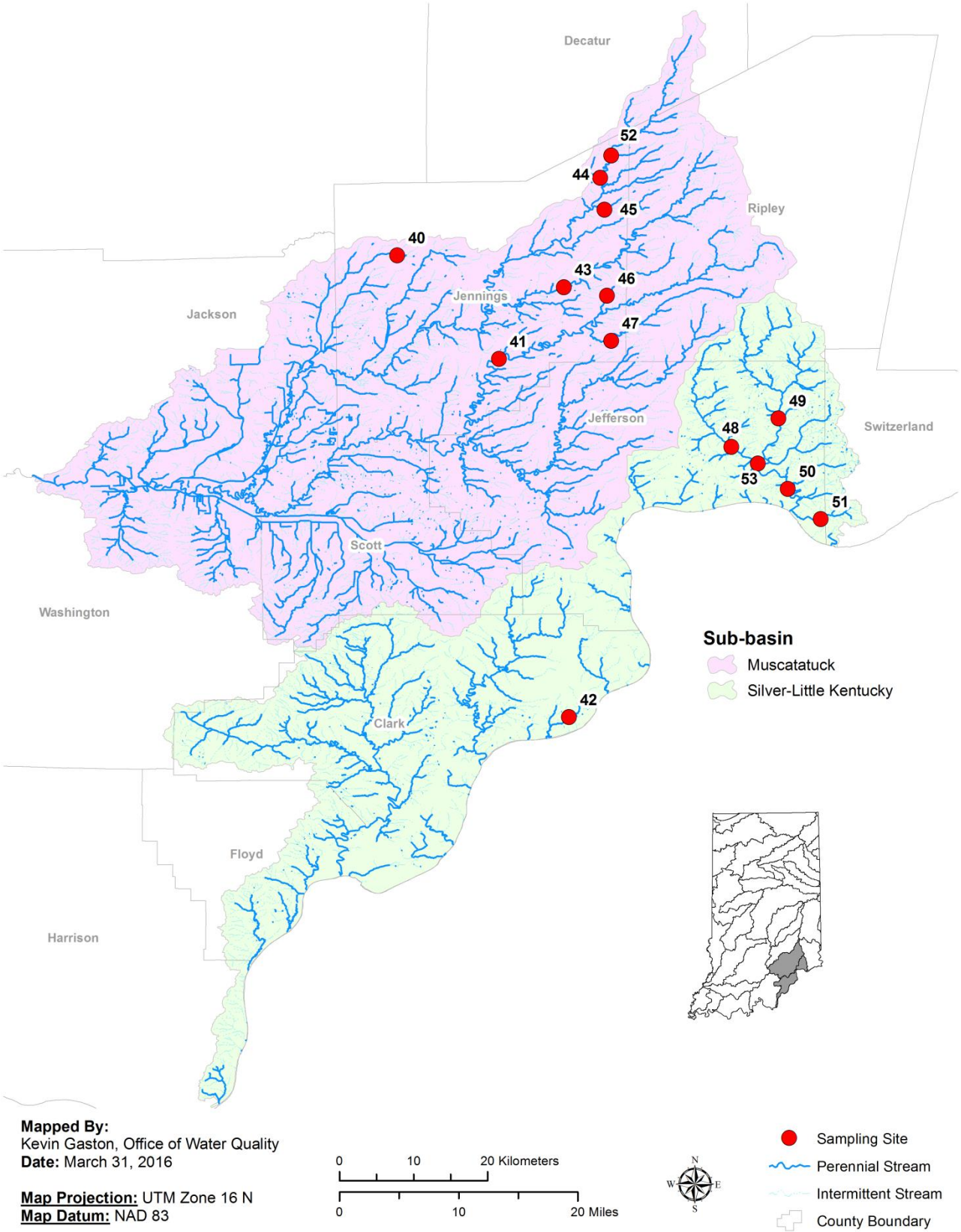


Figure 2. 2016 Reference Sites for the Southern Basin.



6. Specify Tolerable Limits on Decision Errors

Good quality data are essential for minimizing decision error. By identifying errors in the sampling design, measurement, and laboratory for physical, chemical, and biological parameters more confidence can be placed in IBI calibrations and biological threshold determinations as well as aquatic life use assessments.

Decision Error Associated with Reference Site Monitoring

	Actual Status of Sampled Stream Reaches of the Studied Watershed	
WAPB Work Plan Findings	**Stream reach IS supportive of aquatic life use**	**Stream reach IS NOT supportive of aquatic life use**
Stream reach IS supportive of aquatic life use	Stream reach is correctly identified as supporting aquatic life use	**Decision Error (Type 1)**
Stream reach IS NOT supportive of aquatic life use	**Decision Error (Type 2)**	Stream reach is correctly identified as NOT supporting aquatic life use

Site specific aquatic life use assessments include program specific controls to identify the introduction of errors. These controls include water chemistry blanks and duplicates, biological site revisits or duplicates, and laboratory controls through verification of species identifications as described in Field Procedure Manuals (IDEM 2002; Ohio Environmental Protection Agency 2006) and Standard Operating Procedures (SOPs, IDEM 1992a, 1992b, 1992c, 1992d, 1992e, 2010a, 2015a).

The QA/QC process detects deficiencies in the data collection as set forth in the IDEM QAPP for the Indiana Surface Water Quality Monitoring Program (IDEM 2004). The QAPP requires all contract laboratories to adhere to rigorous standards during sample analyses and to provide good quality usable data. Chemists within the WAPB review the laboratory analytical results for quality assurance. Any data which is "Rejected" due to analytical problems or errors will not be used for water quality assessment decisions. Any data flagged as "Estimated" may be used on a case-by-case basis. Criteria for acceptance or rejection of results as well as application of data quality flags is presented in the QAPP, Table D3-1: Data Qualifiers and Flags, pages 130-131. Precision and accuracy goals with acceptance limits for applicable analytical methods are provided in the QAPP, Table A7-1: Precision and Accuracy Goals for Data Acceptability by Matrix, pages 45-47 and Table B2-2: Field Parameters page 81. Further investigation will be conducted in response to consistent "rejected" data in determining the source of error. Field techniques used during sample collection and preparation, along with laboratory procedures will be subject to evaluation by both the WAPB QA Manager and Project Manager in troubleshooting error introduced throughout the entire data collection process. Corrective actions will be implemented once the source of error is determined.

7. Optimize the Design for Obtaining Data

Sampling locations in this project have been selected based on sites that were previously sampled from 2003-2013. Reference sites were least impacted by anthropogenic sources and had good habitat and water chemistry results compared to other sites sampled over the 10-year period. Sampling locations may be near bridges or in remote areas due to being a historical probabilistic site.

Indiana's 2014 CALM (pages 22-23) requires at least three samples in order to complete an assessment for aquatic life use with water chemistry data. Sampling for water chemistry will occur in the spring (April/May), summer (June), and fall (end of August).

Training and Staffing Requirements

Role	Required Training/Experience	Responsibilities	Training References
Project Manager	-Bachelor of Science Degree in biology or other closely related area plus four years of experience in aquatic ecosystems (Master's Degree with two years aquatic ecosystems experience may substitute) - Database experience - Experience in project management and QA/QC procedures	-Establish Project in the Assessment Information Management System (AIMS) II database -Oversee development of Project Work Plan -Oversee entry and QC of field data -Oversee querying of data from AIMS II database to determine results not meeting aquatic life use Water Quality Criteria	-AIMS II Database User Guide - U.S. EPA 2006 Quality Assurance (QA) Documents on developing Work Plans (QAPPs)
Field Crew Chief-Fish or Macroinvertebrate Community Sampling	-Bachelor of Science Degree in biology or other closely related area - At least one year of experience in sampling methodology and taxonomy of aquatic communities in the region -Annually review the Principles and Techniques of Electrofishing -Annually review relevant safety procedures -Annually review relevant SOP documents for field operations	-Completion of field data sheets -Taxonomic accuracy -Sampling efficiency and representation -Voucher specimen tracking -Overall operation of field crew when remote from central office -Adherence to safety and field SOP procedures by crew members -Ensure datasondes calibrated weekly, field sampling equipment is functioning properly, and all equipment loaded into vehicles prior to field sampling activities	-Barbour et al. 1999 -Hydrolab Corporation 2002 -IDEM 1992a, 1992b, 1992c, 1992d, 2002, 2010a, 2010b, 2010c, 2015b -Klemm et al. 1990 -OHEPA 2006 -Plafkin et al. 1989 -Rankin 1995 -Simon and Dufour 2005 -U.S. EPA 1995 -YSI 2002
Field Crew members-Fish or Macroinvertebrate Community Sampling	-Complete hands-on training for sampling methodology prior to field sampling activities -Review the Principles and Techniques of Electrofishing -Review relevant safety procedures -Review relevant SOP documents for field operations	-Follow all safety and SOP procedures while engaged in field sampling activities -Follow direction of Field Crew Chief while conducting field sampling activities	-Barbour et al. 1999 -Hydrolab Corporation 2002 -IDEM 1992a, 1992b, 1992c, 1992d, 2002, 2010a, 2010b, 2010c, 2015b -Klemm et al. 1990 -OHEPA 2006 -Plafkin et al. 1989 -Rankin 1995 -Simon and Dufour 2005 -U.S. EPA 1995 -YSI 2002

Role	Required Training/Experience	Responsibilities	Training References
Field Crew Chief - Water Chemistry and Algal Sampling	-Bachelor of Science Degree in biology or other closely related area -At least one year of experience in sampling methodology -Annually review relevant safety procedures -Annually review relevant SOP documents for field operations	-Field data sheets complete -Sampling efficiency and representation -Overall operation of field crew when remote from central office -Adherence to safety and field SOP procedures by crew members - Ensure datasondes calibrated weekly, field sampling equipment is functioning properly, and all equipment loaded into vehicles prior to field sampling activities	-Hydrolab Corporation 2002 -IDEM 1997, 2002, 2010b, 2010c, 2015a, 2015b -Lowe et al. 2004 -Moulton et al. 2002 -YSI 2002
Field Crew Members - Water Chemistry and Algal Sampling	-Complete hands-on training for sampling methodology prior to field sampling activities -Review relevant safety procedures and SOP documents for field operations	-Follow all safety and SOP procedures while conducting field sampling activities -Follow direction of Field Crew Chief while conducting field sampling activities	-Hydrolab Corporation 2002 -IDEM 1997, 2002, 2010b, 2010c, 2015a, 2015b -Lowe et al. 2004 -Moulton et al. 2002 -YSI 2002
Laboratory Supervisor - Fish or Macroinvertebrate Community Sample Processing	-Bachelor of Science Degree in biology or other closely related area -At least one year of experience in taxonomy of aquatic communities in the region -Annually review relevant safety procedures -Annually review relevant SOP documents for laboratory operations	-Adherence to safety and SOP procedures by laboratory staff -Assist with identification of fish/macroinvertebrate specimens -Verify taxonomic accuracy of samples -Voucher specimen tracking -QC calculations on data sheets, check for completeness -Ensure data are entered into AIMS II correctly	-IDEM 1992a,1992e, 2004, 2008, 2010b, 2010c -AIMS II Database User Guide
Laboratory Staff - Fish or Macroinvertebrate Community Sample Processing	-Complete hands-on training for laboratory sample processing methodology prior to laboratory sample processing activities -Annually review relevant safety procedures and relevant SOP documents for laboratory operations	-Adhere to safety and SOP procedures -Follow Laboratory Supervisor direction while processing samples -Identify fish/ macroinvertebrate specimens -Perform necessary calculations on data, enter field sheets	-IDEM 1992a,1992e, 2004, 2008, 2010b, 2010c -AIMS II Database User Guide

Role	Required Training/Experience	Responsibilities	Training References
Laboratory Supervisor - Water Chemistry and Algal Sample Processing	-Bachelor of Science Degree in biology or other closely related area -Annually review relevant safety procedures -Annually review relevant SOP documents for field operations	-Adherence to safety and SOP procedures by laboratory staff -Identification of diatoms -Completion of laboratory data sheets -Check data for completeness -Perform all necessary calculations on the data -Ensure that data are entered into the AIMS II database	-IDEM 2010b, 2010c, 2015a - Barbour et al. 1999 -Biggs and Kilroy 2000 - H. Ettl, J. Gerloff, H. Heynig, D. Mollenhauer 1986, 1988, 1991 - Prescott 3rd ed. 1970 -Wehr and Sheath eds. 2003 - John, Whitton, and Brookes 2002 - William Vinyard 1979 -AIMS II Database User Guide
Quality Assurance Officer	-Bachelor of Science in chemistry or a related field of study -Familiarity with QA/QC practices and methodologies -Familiarity with the WAPB QAPP and data qualification methodologies	-Ensure adherence to QA/QC requirements of WAPB QAPP -Evaluate data collected by sampling crews for adherence to project Work Plan -Review data collected by field sampling crews for completeness and accuracy -Perform a data quality analysis of data generated by the project - Assign data quality levels based on the data quality analysis -Import data into the AIMS II database -Ensure that field sampling methodology audits are completed according to WAPB procedures	-IDEM 2004, 2012b -U.S. EPA 2006 documentation on QAPP development and data qualification -AIMS II Database User Guide

II. MEASUREMENT/DATA ACQUISITION

Sampling Design and Site Locations

The reference site locations proposed in this project have been selected because they were previously sampled for fish and/or macroinvertebrates with habitat evaluations and some water chemistry results. After evaluating watershed characteristics (land use, pollution sources, road

density, percent impervious surface, etc.) as well as habitat and chemistry results, these reference site locations were considered least impacted by anthropogenic sources.

Site reconnaissance activities will be conducted in-house and through physical site visits. In-house activities include preparation and review of site maps and aerial photographs, potential access routes and property owner searches. Physical site visits include property owner consultations, confirmation and documentation of access routes, and determination of equipment needed to properly sample the site. All information will be recorded on the IDEM Site Reconnaissance Form (Attachment 1) and entered into the AIMS II database. Final coordinates for each site will be determined using a Trimble Juno™ SB handheld Series Global Positioning System (GPS) with an accuracy of 2-5 meters (IDEM 2015b).

Sampling Methods

Water Chemistry

During three discrete sampling events, one team of two staff will collect grab water chemistry samples and record water chemistry field measurements and physical site descriptions on the IDEM Stream Sampling Field Data Sheet (Attachment 2). There will be a minimum of four weeks between samples. All water chemistry sampling procedures will adhere to the Water Quality Surveys Section Field Procedure Manual 2.0 (Field Procedure Manual IDEM 2002, pages 8-14). Water chemistry sampling usually takes 30 minutes to complete for each site, depending on accessibility.

Field Parameter Measurements

Dissolved oxygen (DO), pH, water temperature, specific conductance, and DO percent saturation will be measured with a data sonde during each sampling event regardless of the media type being collected. Measurement procedures and operation of the data sonde shall be performed according to the manufacturers' manuals (Hydrolab Corporation 2002; YSI 2002) and Sections 2.10-2.13 of the Water Quality Surveys Section Field Procedure Manual (IDEM 2002, pages 67-79). Turbidity will be measured with a Hach™ turbidity kit, and the meter number written in the comments under the field parameter measurements. All field parameter measurements and weather codes will be recorded on the IDEM Stream Sampling Field Data Sheet (Attachment 2) with other sampling observations. A digital photo will also be taken upstream and downstream of the site during each sampling event.

Algal Sampling

In addition to standard water chemistry sampling, one team of two staff will collect chlorophyll *a* and pheophytin *a* from the phytoplankton (seston) and periphyton communities during the third round of water chemistry. In order to obtain a representative algal community sample, collection must occur during low/base flow and not directly following a major precipitation event (i.e., a sudden rain event that quickly increases the stream flow above low/base flow, determined either by viewing recent data from the USGS stream flow monitoring gages (available at http://waterdata.usgs.gov/nwis/rt) or by best professional judgment during a visit to the site. Following major weather events, sampling must be postponed for a week to allow the algal communities to return to a representative state. Sampling for an average site that includes all of the above parameters will require approximately 2.5 hours of effort. The Algal Biomass Lab Data Sheet (Attachment 3) and Physical Description of Stream Site Form (Attachment 4) will be used to record information regarding substrates sampled for periphyton and physical parameters of the stream sampling area.

Data analysis by the USGS indicated no correlation between phytoplankton chlorophyll *a* and effects on biological communities in headwater (52 km^2 or less) and wadeable (52 - 2590 km^2) streams (Caskey et al. 2013); therefore, phytoplankton samples will only be collected in streams greater than 2590 km^2 (1000 mi^2) to save resources both in terms of sampling time and costs

associated with chlorophyll *a* analysis. All phytoplankton samples will be collected along a transect in the stream using either the multiple vertical method (if flow >1.5 ft/s) or the grab sample method (gloves must be worn at all times) (IDEM 2016a).

Periphyton samples will be collected from one of three substrate types (in order of preference): epilithic (rocks), epidendric (sticks), or epipsammic (sand). Rocks represent the most stable substrate, which more accurately reflects stream conditions from a specific site, so they are given precedence over sticks and sand. Sand is the most frequently disturbed substrate and therefore least representative of a stable climax algal community. Sand is only collected if rocks and sticks are not present at a site (IDEM 2016a).

Samples will be delivered to the USGS Indiana Algal Biomass Laboratory in Indianapolis and processed within 24 days of collection. Using U.S. EPA Method 445.0, the laboratory will provide measurements for chlorophyll *a* and pheophytin *a* for both seston and periphyton samples.

Laboratory Procedures for Diatom Identification and Enumeration

Many diatom samples will contain large amounts of organic matter as well as diatoms. Therefore, diatom samples will need to be cleaned with acid (or some other strong oxidizing agent) before they are made into permanent mounts (Biggs and Kilroy 2000). We have chosen to use the Hydrogen Peroxide/Potassium Dichromate Oxidase Cleaning Method as described in Barbour et al. 1999. See IDEM 2015a for a description of methods used in Diatom Identification and Enumeration.

Fish Community Sampling

Taxonomic characteristics for possible species encountered in the basin of interest will be reviewed prior to field work through discussion of PowerPoint slides of fish species and laboratory examination of reference fish collections.

Fish community sampling will be performed using various standardized electrofishing methodologies, depending on stream size and site accessibility. Fish assemblage assessments will be performed in a sampling reach of 15 times the average wetted width, with a minimum reach of 50 meters and a maximum reach of 500 meters (Simon and Dufour 2005; U.S. EPA 1995). An attempt will be made to sample all habitat types available within the sample reach to ensure adequate representation of the fish community present at the time of the sampling event. The possible list of electrofishers to be utilized include: the Smith-Root LR-24 or LR-20B Series backpack electrofishers, the Smith-Root model 1.5KVA electrofishing system, the Smith-Root model 2.5 Generator Powered Pulsator (GPP) electrofisher with RCB-6B junction box and rat-tail cathode cable assembled in a canoe (if parts of the stream are not wadeable, the system may require the use of a dropper boom array outfitted in a canoe or possibly a 12-14 foot boat), or, for non-wadeable sites, the Smith-Root model 6a electrofisher assembled in a 16-foot boat (IDEM 1992a, 1992b, 1992c, 1992d).

Sample collections during high flow or turbid conditions will be avoided due to 1) low collection rates which result in non-representative samples and 2) safety considerations for the sampling team. Sample collections during late autumn and seasonal cold temperatures will be avoided due to the lack of responsiveness to the electrical field by some species that can also result in samples that are not representative of the streams fish assemblage (Simon 1990; U.S. EPA 1995).

Fish will be collected using dipnets with fiberglass handles and netting of 1/8-inch bag mesh. Fish collected in the sampling reach will be sorted by species into baskets and buckets. Young-of-the year fish less than 20 millimeters (mm) total length will not be retained in the community sample (Simon 1990; U.S. EPA 1995).

Prior to processing fish specimens and completion of the fish community datasheet, one to two individuals per species will be preserved in 3.7% formaldehyde solution for future reference if there are more than 10 individuals for that species collected in the sampling reach, the specimens can be positively identified, and the individuals for preservation are small enough to fit in a 2000 mL jar. If however, there are few individuals captured or the specimens are too large to preserve, a photo of key characteristics will be taken for later examination. Fish specimens should also be preserved if they cannot be positively identified in the field (especially those that co-occur like the Striped and Common Shiners), individuals that appear to be hybrids or have unusual anomalies, as well as dead specimens that are taxonomically valuable for un-described taxa (like the Red Shiner or Jade Darter), life history studies, or research projects.

Data will be recorded for non-preserved fish on the IDEM Fish Collection Data Sheet (Attachment 5) consisting of the following: number of individuals, minimum and maximum total length (mm), mass weight in grams (g), and number of individuals with deformities, eroded fins, lesions, tumors, and other anomalies (DELTs). Once the data have been recorded, specimens will be released within the sampling reach if possible. Data will be recorded for preserved fish specimens following taxonomic identification in the laboratory.

Macroinvertebrate Sampling

Aquatic benthic macroinvertebrate samples are collected using a modification of the U.S. EPA Rapid Bioassessment Protocol multi-habitat (MHAB) approach using a D-frame dip net with 500 µm mesh (Plafkin et al. 1989; Barbour et al. 1999; Klemm et al. 1990; IDEM 2010a). The IDEM MHAB approach (IDEM 2010a) is composed of a 1-minute “kick” sample within a riffle or run and a 50 meter “sweep” sample of shoreline habitats. The 50 meter length of riparian corridor that is sampled at each site will be defined using a rangefinder or GPS unit. If the stream is too deep to wade, a boat will be used to sample the 50 meter zone along the shoreline that has the best available habitat. The 1-minute “kick” and 50 meter “sweep” samples are combined in a bucket of water which will be elutriated through a U.S. standard number 35 (500 µm) sieve a minimum of five times so that all rocks, gravel, sand, and large pieces of organic debris are removed from the sample. The remaining sample is then transferred from the sieve to a white plastic tray where the collector (while still on-site) will conduct a 15-minute pick of macroinvertebrates at a single organism rate with an effort to pick for maximum organism diversity through turning and examination of the entire sample in the tray. The resulting picked sample will be preserved in 70% isopropyl alcohol and returned to the laboratory for identification at the lowest practical taxonomic level (usually genus or species level, if possible) and evaluated using the MHAB macroinvertebrate IBI. Before leaving the site, an IDEM OWQ Macroinvertebrate Header Form (Attachment 6) will be completed for the sample.

In addition to the standard MHAB method of macroinvertebrate collection, three other macroinvertebrate sampling methods will be employed at each reference site. These three additional sampling methods were developed to validate components of the MHAB method, specifically the 15-minute field pick and use of a 50 meter sample zone instead of a sample zone that is a multiple of the stream width (i.e. 15 times the streams wetted width). These methods were first employed at randomly selected sites in the 2013 and 2014 sample seasons (IDEM 2014c); analysis of these earlier samples may prove that a particular alternate method is superior, at which point only that method and the MHAB method will be used at reference sites. The three alternate methods are:

1. Keeping the “un-picked” remainder of the MHAB sample after completion of the 15-minute pick
2. Collection of three “jabs” taken with a D-frame dipnet on each of 10 equally spaced transects (30 jabs total) located on a stretch of stream equal to 15 times the wetted width of the stream at the site location
3. Collection of two 0.25 square meter “kick” samples taken with a 0.5 meter wide bottom kick net, collected from alternating thirds of each transect line (total of five square meters of stream substrate).

At three reference sites, an additional duplicate set of all four sampling methods will be collected. The samples collected in 2016 will increase the total number of samples collected for the methods comparison study to 80 with 16 sets of duplicate samples.

Habitat Assessments

Habitat assessments will be completed immediately following macroinvertebrate and fish community sample collections at each site using a slightly modified version of the Ohio Environmental Protection Agency (OHEPA) QHEI, 2006 edition (Rankin 1995; OHEPA 2006). A separate Qualitative Habitat Evaluation Index (QHEI) (Attachment 7) must be completed for these two media types since the sampling reach length may differ (i.e. 50 meters for macroinvertebrates and between 50 and 500 meters for fish) (IDEM 2016b).

Analytical Methods

Table 4 lists the field parameters with their respective test method and IDEM quantification limits. Table 5 lists the algal parameters with test method and USGS quantification limits. Table 6 shows water chemistry sample container, preservative, and holding time requirements (all samples iced to 4 Degrees Celsius (°C). Table 7 lists numerous parameters (priority metals, anions/physical, and nutrients/organic) with their respective test methods, IDEM reporting limits, and contract laboratory reporting limits.

Diatoms will be collected in the field according to protocols described in Moulton et al. 2002 with a slight modification as mentioned in Section II Algal Sampling. MEASUREMENT/DATA ACQUISITION. Several diatom taxonomic references are used to aid in the identification process. The list of references that will be utilized by branch staff can be found in the technical standard operating procedure for “Processing and Identification of Diatom Samples”, listed as Appendix 4 (IDEM 2015a).

Table 4. Field Parameters showing method and IDEM quantification limit.

Parameters	Method (SM=Standard Method)	IDEM Quantification Limit
Dissolved Oxygen (data sonde optical)	ASTM D888-09	0.05 mg/L
Dissolved Oxygen (data sonde)	SM 4500-OG	0.03 mg/L
Dissolved Oxygen (Winkler Titration)	SM 4500-OC [1]	0.20 mg/L
Dissolved Oxygen % Saturation (data sonde optical)	ASTM D888-09	0.05 %
Dissolved Oxygen % Saturation (data sonde)	SM 4500-OG	0.01 %
pH (data sonde)	EPA 150.2	0.10 S.U.
pH (field pH meter)	SM 4500H-B [1]	0.10 S.U.
Specific Conductance (data sonde)	SM 2510B	1.00 µmho/cm
Temperature (data sonde)	SM 2550B(2)	0.1 °C
Temperature (field meter)	SM 2550B(2) [1]	0.1 °C
Turbidity (data sonde)	SM 2130B	0.02 NTU [2]
Turbidity (Hach™ turbidity kit)	EPA 180.1	0.05 NTU [2]

[1] Method used for Field Calibration Check
[2] NTU = Nephelometric Turbidity Unit(s)

Table 5. Algal Parameters showing method and USGS quantification limit.

Algal Parameter	Method	USGS Quantification Limit
Seston Chlorophyll *a* - Suspended	U.S. EPA 445.0	0.30 µg/L
Seston Pheophytin *a* - Suspended	U.S. EPA 445.0	0.30 µg/L
Periphyton Chlorophyll *a* - Attached	U.S. EPA 445.0	0.30 µg/m^2
Periphyton Pheophytin *a* - Attached	U.S. EPA 445.0	0.30 µg/m^2

Table 6. Water Chemistry Sample Container, Preservative, and Holding Time Requirements.

Parameter	Container	Preservative	Holding
[1]Alkalinity as $CaCO_3$*	1 L, plastic, narrow mouth	None	14 days
[2]Ammonia-N**	1 L, plastic, narrow mouth	H_2SO_4 < pH 2	28 days
Chloride*	1 L, plastic, narrow mouth	None	28 days
Chemical Oxygen Demand**	1 L, plastic, narrow mouth	H_2SO_4 < pH 2	28 days
Hardness (as $CaCO_3$*) Calculated	1 L, plastic, narrow mouth	HNO_3 < pH 2	6 months
Metals (Total & Dissolved)	1 L, plastic, narrow mouth	HNO_3 < pH 2	6 months
Nitrate + Nitrite-N**	1 L, plastic, narrow mouth	H_2SO_4 < pH 2	28 days
Total Phosphorus**	1 L, plastic, narrow mouth	H_2SO_4 < pH 2	28 days
Solids (All Forms)*	1 L, plastic, narrow mouth	None	7 days
Sulfate*	1 L, plastic, narrow mouth	None	28 days
Total Kjeldahl Nitrogen**	1 L, plastic, narrow mouth	H_2SO_4 < pH 2	28 days
Total Organic Carbon**	1 L, plastic, narrow mouth	H_2SO_4 < pH 2	28 days

[1]General chemistry includes all parameters noted with an *.
[2]Nutrients include all parameters noted with a **.

Table 7. Water Chemistry Parameters, Test Method, IDEM and Laboratory Reporting Limits.

Priority Metals					
Parameter	Total	Dissolved	Test Method	IDEM Reporting Limit (µg/L)	Pace Laboratory Reporting Limit (µg/L)
Aluminum	☒	☒	EPA 200.7	150	20
Antimony	☒	☒	EPA 200.8	1	0.5
Arsenic	☒	☒	EPA 200.8	5	2.5
Calcium	☒	☐	EPA 200.7	40	40
Cadmium	☒	☒	EPA 200.8	2	1
Chromium	☒	☒	EPA 200.8	3	1.5
Copper	☒	☒	EPA 200.8	2	1
Lead	☒	☒	EPA 200.8	2	1
Magnesium	☒	☐	EPA 200.7	95	100
Nickel	☒	☒	EPA 200.8	1.5	0.75
Selenium	☒	☒	EPA 200.8	4	2
Silver	☒	☒	EPA 200.8	0.3	0.3
Zinc	☒	☒	EPA 200.8	6	6

Anions/Physical			
Parameter	Pace Test Method	IDEM Reporting Limit (mg/L)	Pace Laboratory Reporting Limit (mg/L)
Alkalinity (as CaCO3)	EPA 310.2	10	10
Total Solids	SM2540B	1	10
Total Suspended Solids	SM2540D	1	1
Dissolved Solids	SM2540C	10	10
Sulfate	EPA 300.0	0.05	0.35
Chloride	EPA 300.0	1	1
Hardness (as CaCO3) by calculation	SM2340B	0.4	1

Nutrients/Organic			
Parameter	Pace Test Method	IDEM Reporting Limit (mg/L)	Pace Laboratory Reporting Limit (mg/L)
TKN	SM4500N(Org)	0.03	0.3
Ammonia-N	SM4500NH3-G	0.01	0.1
Nitrate+Nitrite-N	EPA 353.2	0.05	0.01
Total Phosphorus	EPA 365.1	0.01	0.05
Total Organic Carbon (TOC)	SM5310C	1	1
Chemical Oxygen Demand (COD)	EPA 410.4	3	10

Quality Control and Custody Requirements

Quality assurance protocols will follow part B5 of the WAPB QAPP (IDEM 2004 page 119-121).

Water Chemistry Data

Sample bottles and preservatives used will be certified for purity by the manufacturer. Sample collection for each parameter, preservatives, and holding times will adhere to U.S. EPA requirements (U.S. EPA 2007). See Table 6.

- Field duplicates and matrix spike/matrix spike duplicates (MS/MSD) shall be collected at the rate of one per sample analysis set or one per every 20 samples, whichever is greater.
- Field blank samples using ASTM D1193-91 Type I water will be taken at a rate of one per sample analysis set or one per every 20 samples, whichever is greater.

The IDEM OWQ Chain of Custody Form (Attachment 8) and the 2016 Reference Sites Water Sample Analysis Request Form (Attachment 9) accompanies each sample set through the analytical process.

Algal Community Data

Excessive algal conditions will be recorded by staff if an algal bloom is observed on the water's surface or in the water column. Staff are not calibrated on this rating (i.e. the decision as to the severity of the bloom is based on best professional judgement), but an algal mat on the surface of the water or a bloom that gives the water the appearance of green paint would be justification for a decision of excessive algal conditions. To decrease the potential for cross contamination and bias of the algal samples, all equipment that has come in contact with the sample will be cleaned with detergent and rinsed with ASTM D1193-91 Type III water after sampling has been completed at a given site. All sample labels must be accurately and thoroughly completed, including AIMS II database sample numbers, date, stream name, and sampling location. The sample collection portion of the Chain of Custody forms will be completed in the field. The form will be completed when samples are transferred to the laboratory. Upon arrival to the laboratory, samples will be checked in by the laboratory manager. For the diatom samples, there will be another Chain of Custody form to document when the sample is removed from storage to be processed and made into a permanent mount (IDEM 2015a).

Methods and quantification limits for chlorophyll *a* and pheophytin *a* can be viewed in Table 4. All samples collected for chlorophyll *a* and pheophytin *a* determination will be processed by the USGS Indiana Algal Biomass Laboratory (Indianapolis, Indiana) following the specifications set in Joint Funding Agreement EDS# A305-3-109 (IDEM 2013) and amendment (IDEM 2015c). Blank filters will be run for periphyton and seston chlorophyll *a*. All chlorophyll *a* and pheophytin *a* filters will be processed in triplicate for QC purposes. Ten percent of these replicate field samples will be analyzed at the USGS National Water Quality Laboratory in Arvada, Colorado.

Quality control of the diatom sampling, enumeration, and identification project will be documented by QC checks of both field and laboratory data. See IDEM 2015a for description of quality assurance/ quality control protocols used in Diatom Identification and Enumeration. Ten percent of diatom samples will be verified by the Department of Biological and Environmental Sciences of Georgia College and State University (Milledgeville, Georgia) following the specifications set forth in IDEM 2015a and IDEM 2014c.

Fish Community Data

Replicate fish community sampling will be performed at a rate of 10 percent of the total fish community sites sampled, 3 sites chosen using a random numbers table for the project (IDEM 1992a; U.S. EPA 1995). Replicate sampling will be performed with at least 2 weeks of recovery

between the initial and replicate sampling events. The fish community replicate sampling and habitat assessment will be performed with either a partial or complete change in field team members (U.S. EPA 1994; U.S. EPA 1995). The resulting IBI and QHEI total score between the initial visit and the revisit will be used to evaluate precision. The IDEM Biological Samples Field Chain of Custody Form is used to track samples from the field to the laboratory (Attachment 10). Fish in the laboratory may be verified by regionally recognized non-IDEM freshwater fish taxonomists (i.e. Brant Fisher, Indiana DNR). All data: 1) are checked for completeness, 2) have calculations performed, 3) are entered into the AIMS II database and 4) rechecked for data entry errors.

Macroinvertebrate Community Data

Replicate macroinvertebrate field samples will be collected at a rate of 10 percent of the total macroinvertebrate community sites sampled, approximately 3 for the project. The macroinvertebrate community replicate sample and habitat assessment will be performed by the same team member who performed the original sample, immediately after the initial sample is collected. This will result in a precision evaluation based on a 10 percent replicate of samples collected. The IDEM Biological Samples Field Chain of Custody Form is used to track samples from the field to the laboratory (Attachment 10). Laboratory identifications and QA/QC of taxonomic work is maintained by the laboratory supervisor, Macroinvertebrate Community Program Manager.

Field Parameter Measurements/Instrument Testing/Calibration

The data sonde will be calibrated prior to each week's sampling (IDEM 2002). The DO component of the calibration procedure will be conducted using the air calibration method (IDEM 2002, pp. 74-75). Calibration results and drift values will be recorded and stored in log books located in the calibration laboratories at the Shadeland facility. The drift value is the difference between two successive calibrations. Field parameter calibrations will conform to the procedures as described in the instrument users' manuals (Hydrolab Corporation 2002; YSI 2002). The unit will be field checked for accuracy once during the week by comparison with a Winkler DO test (IDEM 2002, pp. 64-66), as well as Hach™ turbidity, pH, and temperature meters. Weekly calibration verification results will be recorded on the field calibrations portion of the Stream Sampling Field Data Sheet (Attachment 2) and entered into the AIMS II database. A Winkler DO test will also be conducted at sites where the DO concentrations detected using a data sonde are 4.0 mg/L or less.

Field Analysis Data

In-situ water chemistry field data will be collected in the field using calibrated or standardized equipment. Calculations may be done in the field or later at the office. Analytical results, which have limited QC checks, are included in this category. Detection limits have been set for each analysis and are listed in Table 4. Quality control checks (such as duplicate measurements, measurements of a secondary standard, or measurements using a different test method or instrument) which are performed on field or laboratory data are usable for estimating precision, accuracy, and completeness for the project.

Algal Community Data

Equipment required for the collection of periphyton include: a toothbrush, cloth measuring tape, petri dish top, spatula, stencil brush, small hobby knife with a chisel blade, a dissection probe, a modified syringe with a rubber O-ring attached, Nalgene© high density polyethylene (HDPE) plastic 250 mL sample bottles, plastic bins, and a unitary wash bottle filled with tap water. None of this equipment requires calibration. Equipment has been field tested to ensure its capability of appropriately removing periphyton from different types of substrate (rocks, sticks, sand/silt).

Laboratory equipment that will be used for the preparation of permanent diatom mounts include: hot plate, fume hood, centrifuge, glass beakers, centrifuge tubes, glass microscope slides, microscope cover glasses, micropipette, and micropipette tips. The micropipette was purchased new and came with a calibration certificate as proof that it was calibrated at the factory. Other than the micropipette, none of the laboratory equipment requires calibration. The micropipette will be checked and recalibrated as necessary according to manufacturer's specifications.

A Nikon© DIC microscope and Nikon© Elements D camera and imaging system will be used for identification and enumeration of diatoms. Branch staff calibrated the ocular reticle in the microscope. The ocular reticle was calibrated at each magnification with a stage micrometer.

III. ASSESSMENT/OVERSIGHT

Field and laboratory performance and system audits will be conducted to ensure good quality data. The field and laboratory performance includes precision measurements by relative percent difference (RPD) of field and laboratory duplicate (IDEM 2004, pp. 41, 45-46), accuracy measurements by percent of recovery of MS/MSD samples analyzed in the laboratory (IDEM 2004, pp. 43, 45-46), and completeness measurements by the percent of planned samples that are actually collected, analyzed, reported, and usable for the project (IDEM 2004, p. 43).

Field audits will be conducted to ensure that sampling activities adhere to approved SOPs. Audits are systematically conducted by WAPB Quality Assurance staff to include all WAPB personnel that engage in field sampling activities. WAPB field staff involved with sample collection and preparation will be evaluated by QA staff trained in the associated sampling SOPs, and in the processes related to conducting an audit. QA staff will produce an evaluation report documenting each audit for review by those field staff audited, as well as WAPB management. Corrective actions will be communicated to, and implemented by, field staff as a result of the audit process (IDEM 2004, p. 126).

Data Quality Assessment Levels

The samples and various types of data collected by this program are intended to meet the quality assurance criteria and Data Quality Assessment (DQA) Levels as described in the WAPB QAPP (IDEM 2004, pp. 128-129).

IV. DATA VALIDATION AND USABILITY

Quality assurance reports to management and data validation and usability are also important components of the QAPP which ensures good quality data for this project. A quality assurance audit report will be submitted to the QA Manager and Project Manager for review for this project should problems arise and need to be investigated and corrected. Data are reduced (converted from raw analytical data into final results in proper reporting units), validated (qualified based on the performance of field and laboratory QC measures incorporated into the sampling and analysis procedures), and reported (described so as to completely document the calibration, analysis, QC measures, and calculations). These steps allow users to assess the data to ensure it meets the project data quality objectives.

Quality Assurance/Data Qualifiers and Flags

The various data qualifiers and flags that will be used for quality assurance and validation of the data are found on pages 130-131 of the WAPB QAPP (IDEM 2004).

Data Usability

The environmental data collected and its usability are qualified and classified into one or more of the four categories: Enforcement Capable Results, Acceptable Data, Estimated Data, and Rejected Data as described on page 130 of the WAPB QAPP (IDEM 2004).

Data collected in 2016 will be recorded in the AIMS II database and presented in three compilation summaries:

- A general compilation of the 2016 Reference Site field and water chemistry data prepared for use in the Indiana Integrated Water Monitoring and Assessment Report.
- A database report format containing biological results and habitat evaluations, which will be produced for inclusion in the Integrated Report as well as individual site folders.
- Laboratory bench sheets of the species taxa names and enumerations of all diatoms collected.

All data and reports will be made available to public and private entities that find the data useful.

Laboratory and Estimated Cost

Laboratory analysis and data reporting for this project will comply with the WAPB QAPP (IDEM 2004), Request for Proposals (RFP) 12-48 (IDEM 2012a), and the OWQ Quality Management Plan (IDEM 2012b). Analytical tests on the water chemistry parameters outlined in Table 7 will be performed by Pace Analytical Services (formerly Heritage Environmental) in Indianapolis, Indiana. Algal samples will be collected by IDEM staff. Chlorophyll *a* and pheophytin *a* will be analyzed by the USGS Indiana Algal Biomass Laboratory, Indianapolis, Indiana. Diatom identification and enumeration will be performed by IDEM staff. Diatom identification and enumeration will be performed by IDEM staff and/or an outside contractor. The Department of Biological and Environmental Sciences, Georgia College and State University will be verifying diatom taxa from ten percent of the sites sampled. All fish and macroinvertebrate samples will be collected and analyzed by IDEM staff. The anticipated budget for laboratory cost for the project is outlined in Table 8.

Table 8. Total Estimated Laboratory Cost for the Project.

Analysis	Laboratory	Estimated Cost
Water Chemistry	Pace Analytical Services (formerly Heritage Environmental) 7726 Moller Road. Indianapolis, Indiana 46268	$28,796
Algal Biomass	USGS Indiana Algal Biomass Laboratory 5957 Lakeside Blvd. Indianapolis, Indiana 46278	$5,375
Diatom Verification	Department of Biological and Environmental Sciences Georgia College and State University 320 S. Wayne St. Milledgeville, GA 31061	$750
	Total	$34,921

Table 9. Personnel Safety and Reference Manuals

Role	Required Training/Experience	Training References	Training Notes
All Staff that Participate in Field Activities	-Basic First Aid and Cardio-Pulmonary Resuscitation (CPR)	-A minimum of 4 hours of in-service training provided by WAPB (IDEM 2010b)	-Staff lacking 4 hours of in-service training or appropriate certification will be accompanied in the field at all times by WAPB staff that

Role	Required Training/Experience	Training References	Training Notes
			meet Health and Safety Training requirements
	-Personal Protective Equipment (PPE) Policy	-IDEM 2008	
	-Personal Flotation Devices (PFD)	-February 29, 2000 WAPB internal memorandum regarding use of approved PFDs	-When working on boundary waters as defined by Indiana Code (IC) 14-8-2-27 or between sunset and sunrise on any waters of the state, all personnel in the watercraft must wear a high intensity whistle and Safety of Life at Sea (SOLAS) certified strobe light.

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*This document may be inspected at the Watershed and Assessment Branch office, located at 2525 North Shadeland Avenue, Indianapolis, IN.

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Kayla Werbianskyj	IDEM/OWQ/WAPB/Targeted Monitoring Section

Attachment 1. IDEM Site Reconnaissance Form.

IDEM

Site Reconnaissance Form

EPA Site Identifier	Rank
Recon #: Trip #:	

Site Number: Stream: County:

Location Description:

Reconnaissance Data Collected

Recon Date | Crew Members

Avg. Width (m) | Avg. Depth (m) | Max. Depth (m) | Nearest Town

Water Present? ☐ | Site Wadeable? ☐ | Riffle/Run Present? ☐ | Road/Public Access Possible? ☐

Site Impacted by Livestock? ☐ | Collect Sediment? ☐ | Gauge Present? ☐

Landowner/Contact Information

First Name | Last Name

Street Address

City | State | Zip

Telephone | E-Mail Address

Pamphlet Distributed? ☐ | Please Call in Advance? ☐ | Results Requested? ☐

Rating, Results, Comments, and Planning

Site Rating By Category (1=easy, 10=difficult)
Access Route
Safety Factor
Sampling Effort

Reconnaissance Decision

Pre-Recon
Recon in process
Approved Site
No, Landowner denied access
No, Dry
No, Stream channel missing
No, Physical barriers
No, Impounded stream
No, Marsh/Wetland
No, Bridge gone or not accessible
No, Unsafe due to traffic or location
No, Site impacted by backwater
No, Other

Equipment Selected

Circle Equipment Needed

Backpack
Boat
Towbarge
Longline
Scanoe
Seine
Weighted Handline
Waders
Gill Net

Comments

Sketch of Stream & Access Route – Indicate Flow, Direction, Obstacles, & Land Use (Use Back of Page, if Necessary)

Attachment 2. IDEM Stream Sampling Field Data Sheet.

Stream Sampling Field Data Sheet

Analysis Set #	EPA Site ID	Rank

Sample #	Site #	Sample Medium	Sample Type	Duplicate Sample #

Stream Name:		River Mile:		County:	

Site Description:

Survey Crew Chief	Sample Collectors 1	2	3	4	Sample Collected Date	Time	Hydrolab #	Water Depth/Gage Ht (ft)	Water Flow (cf/sec)	Flow Estimated?	Algae?	Aquatic Life?
										☐	☐	☐

Sample Taken?	Aliquots	Water Flow Type	Water Appearance	Canopy Closed %
☐ Yes ☐ No; Frozen	☐ 1 ☐ 2 ☐ 3 ☐ 4	☐ Riffle ☐ Dry ☐ Stagnant	☐ Clear ☐ Green ☐ Sheen	☐ 0-20% ☐ 60-80%
☐ No; Stream Dry ☐ No; Other	☐ 6 ☐ 8 ☐ 12 ☐ 24	☐ Pool ☐ Run ☐ Flood	☐ Murky ☐ Black ☐ Other	☐ 20-40% ☐ 80-100%
☐ No; Owner refused Access	☐ 48 ☐ 72 ☐ As-Flow	☐ Glide ☐ Eddy ☐ Other	☐ Brown ☐ Gray (Septic/Sewage)	☐ 40-60%

Special Notes:

Field Data:

Date (m/d/yy)	24-hr Time (hh:mm)	D.O. (mg/l)	pH	Water Temp (°C)	Spec Cond (µohms/cm)	Turbidity (NTU)	% Sat.	Chlorine (mg/l)	Chloride (mg/l)	Chlorophyll (mg/l)	Weather Codes SC	WD	WS	AT
Comments														
Comments														
Comments														
Comments														
Comments														
Comments														

Measurement Flags	
<	< Min. Meter Measurement
>	> Max. Meter Measurement
E	Estimated (See Comments)
R	Rejected (See Comments)

Weather Code Definitions

SC Sky Conditions		WD Wind Direction	WS Wind Strength	AT Air Temp
1 Clear	8 Rain	00 North (0 degrees)	0 Calm	1 < 32
2 Scattered	9 Snow	09 East (90 degrees)	1 Light	2 33-45
3 Partly	10 Sleet	18 South (180 degrees)	2 Mod./Light	3 46-60
4 Cloudy		27 West (270 degrees)	3 Moderate	4 61-75
5 Mist			4 Mod./Strong	5 76-85
6 Fog			5 Strong	6 > 86
7 Shower			6 Gale	

Field Calibrations:

Date (m/d/yy)	Time (hh:mm)	Calibrator Initials	Calibrations Type	Meter #	Value	Units

Calibration Type
pH
DO
Turbidity

Preservatives/Bottle Lots:

Group: Preservative	Preservative Lot #	Bottle Type	Bottle Lot #

Groups: Preservatives		Bottle Types	
GC	General Chemistry: Ice	2000P	2000mL Plastic, Narrow Mouth
Nx	Nutrients: H2SO4	1000P	1000mL Plastic, Narrow Mouth
Metals	Metals: HNO3	500P	500mL Plastic, Narrow Mouth
CN	Cyanide: NaOH	250P	250mL Plastic, Narrow Mouth
O&G	Oil & Grease: H2SO4	1000G	1000mL Glass, Narrow Mouth
Toxics	Toxics: Ice	500G	500mL Glass, Wide Mouth
EcoII	Bacteriology: Ice	250G	250mL Glass, Wide Mouth
VOA	Volatile Organics: HCl & Thiosulfate	125G	125mL Glass, Wide Mouth
Pest	Pesticides: Ice	40GV	40mL Glass Vial
Phen	Phenols: H2SO4	120PB	120ml Plastic (Bacteria Only)
Sed	Sediment: Ice	1000PF	1000mL Plastic, Corning Filter
Gly	Glyphosate: Thiosulfate	500PF	500mL Plastic, Corning Filter
Hg	Mercury(1631): HCl	60P	60mL Plastic
CrS	Chromium VI(1636): NaOH	250T	250mL Teflon
MeHg	Methyl Mercury(1630): HCl	500T	500mL Teflon
		125T	125mL Teflon

Data Entered By: _____ QC1: _____
QC2: _____

Stream Sampling Field Data Sheet

Attachment 3. IDEM Algal Biomass Lab Data Sheet.

Algal Biomass Lab Datasheet

Sample #	Site	Stream

Supporting Site Information

Traditional Forestry % Closed Canopy: ☐ <=10m ☐ >10m (Measure center only if width <=10m, record to nearest whole percent)

	North	East	South	West	Average x 1.04 =
Left Bank					
Center					
Right Bank					
Total %CC (Average from above, or Center only = %CC)				100 - %CC	

Phytoplankton Information

Sampling Method: ☐ Grab Sample (Dip) ☐ Multiple Verticles Number of Verticles:

Chlorphyll A	Blank	Filter 1	Filter 2	Filter 3	Filter 4
Sample Time					
Sample Volume (mL)					

Periphyton Information

Periphyton Habitat: ☐ Epilithic (Area-Scape) ☐ Epidendric (Cylinder Scrape) ☐ Epipsammic (Petri Dish)

Diatom Sample Collected: ☐ Yes ☐ No | Diatom Volume: mL | Formalin Volume: mL | Slurry Volume mL

Chlorphyll A	Blank	Filter 1	Filter 2	Filter 3	Filter 4
Sample Time					
Sample Volume (mL)					

Periphyton Area Calculation

Cylinder Scrape

Snag #	Length (cm)(L)	Circumference U_1	U_2	U_3	U	Area (L * U)
1						
2						
3						
4						
5						
				Total Area (cm²)		

Area Scrape (Using SG-92)

Rock#	1	2	3	4	5
Area (cm²)	7.38	7.38	7.38	7.38	7.38
Total (cm²)	36.9				

Petri Dish

Number of Discrete Samples (n):	
Total Area of One Sampler (a):	19.01 cm²
Total Sample Area (n * a):	

Stream Discharge / Rainfall Information

Nearest USGS Gage Site: ☐ Upstream ☐ Downstream ☐ No USGS Gage Near

River miles from site:	Discharge CFS at sampling: CFS
Gage location:	Discharge days since 50% flow exceeded: days
Rainfall data source: ☐ NOAA ☐ CoCoRaHS ☐ Indiana State Climate Office ☐ USGS gage rain gauge ☐ Other:	
Total precipitation at sampling: in. on date:	Cumulative rain 7 days previous to sampling: in.
Rain station location, county:	Inches since last rainfall previous to sampling: in. Days since last rainfall previous to sampling: days

Identifier	Date	Reviewer 1	Date	Reviewer 2	Date	Notes:
		☐ Review 1 Completed		☐ Review 2 Completed		

Attachment 4. IDEM Physical Description of Stream Site Form (front).

Revised 4/20/12

Probabilistic Monitoring Section Physical Description of Stream Site

Stream :_____ **AIMS #**_____**Program #:**_____

Date: _____ **Time:** _____ **Crew Chief:**_____**Crew**_____

General Stream Description:

_____
_____
_____
_____
_____

Characteristics at the site and immediately upstream (check All that apply).

Outer Riparian Zone L R	Inner Riparian Zone L R	L.Width(m)	R.Width(m)
☐ ☐ Agricultural Row crop	☐ ☐ Agricultural Rowcrop	_____	_____
☐ ☐ Agricultural Pasture	☐ ☐ Agricultural Pasture	_____	_____
☐ ☐ Devoid of Vegetation	☐ ☐ Devoid of Vegetation	_____	_____
☐ ☐ Fallow	☐ ☐ Fallow	_____	_____
☐ ☐ Forested	☐ ☐ Forest	_____	_____
☐ ☐ Residential	☐ ☐ Residential	_____	_____
☐ ☐ Commercial/Industrial	☐ ☐ Commercial/Industrial	_____	_____
☐ ☐ Weeds and Scrub	☐ ☐ Treeline	_____	_____
☐ ☐ Other _____	☐ ☐ Weeds and Scrub		
	☐ ☐ Other _____	_____	_____

Flow above site	Flow at site	Substrate (if visable)
☐ Riffle	☐ Riffle	☐ Cobble
☐ Pool	☐ Pool	☐ Boulder
☐ Eddy	☐ Eddy	☐ Sand
☐ Run	☐ Run	☐ Muck
☐ Glide	☐ Glide	☐ Silt
☐ Other _____	☐ Other _____	☐ Gravel
_____	_____	☐ Bedrock
_____	_____	☐ Other _____

Characteristics at site and immediately upstream (check ONE).

Water Description	Sinuosity of Channel	Discharge Pipe Present
☐ Clear	☐ High	☐ No
☐ Grey (Septic)	☐ Moderate	☐ Yes
☐ Murky	☐ Low	If yes, Effluent Flowing?
☐ Black	☐ Channelized	☐ No
☐ Brown		☐ Yes
☐ Green		Description of Effluent _____
☐ Other _____		_____

Continued on back

Attachment 4. IDEM Physical Description of Stream Site Form (back).

Revised 4/20/12

Stream Bank

Functional Slope: L R	Bank Erosion: L R	
☐☐ 0-30°	☐☐ Low	Percent Canopy Closed: _____
☐☐ 31-50°	☐☐ Moderate	Stream Stage 1-5 (Low-High): _____
☐☐ 51-70°	☐☐ High	Velocity of Stream 1-5 (Slow-Fast): _____
☐☐ 71-90°		

Visible Stream Degradation? ☐ Yes ☐ No

Description:_____

Aquatic Life Observed? ☐ Yes ☐ No

Description: _____

Algae Observed? ☐ Yes ☐ No

Description: _____

Rooted Macrophytes Observed? ☐ Yes ☐ No

Description: _____

Additional Comments:

_____
_____
_____
_____

Follow Up Date:_____Time:_____Crew Chief:_____Crew:_____
_____
_____
_____
_____

Follow Up Date:_____Time:_____Crew Chief:_____Crew:_____
_____
_____
_____
_____

Photography Date:_____ Time:_____ Number(s):_____; _____;_____

Notes (include items relevant for determining scale – items of known measurement, etc.)
_____
_____
_____

Attachment 5. IDEM Fish Collection Data Sheet (front).

IDEM
OWQ-WATERSHED ASSESSMENT AND PLANNING BRANCH

Event ID______________ Voucher jars_______ Unknown jars________ Equipment_______________ Page _____ of ______
Voltage_______ Time fished (sec)________ Distance fished (m)_________ Max. depth (m)_________ Avg. depth (m)________
Avg. width (m)______ Bridge in reach_____ Is reach representative_____ If no, why_______________________________________
Elapsed time at site (hh:mm)____:____ Comments____
____
____

Museum data: Initials__________ ID date__________ Jar count__________ Fish Total__________

Coding for Anomalies: D – deformities E – eroded fins L – lesions T – tumor M – multiple DELT anomalies O – other (A – anchor worm C – leeches W – swirled scales Y – popeye S – emaciated F – fungus P – parasites) H – heavy L – light (these codes may be combined with above codes)

TOTAL # OF FISH				WEIGHT (s) (mass g)				(length mm)	ANOMALIES					
								Min length	D	E	L	T	M	O
								Max length						
V		P												
								Min length	D	E	L	T	M	O
								Max length						
V		P												
								Min length	D	E	L	T	M	O
								Max length						
V		P												
								Min length	D	E	L	T	M	O
								Max length						
V		P												
								Min length	D	E	L	T	M	O
								Max length						
V		P												
								Min length	D	E	L	T	M	O
								Max length						
V		P												

MKM: Rev/February 19, 2014

Attachment 5. IDEM Fish Collection Data Sheet (back).

Event ID_______________ Page _____ of ______

								Min length	D	E	L	T	M	O
								Max length						
V		P												
								Min length	D	E	L	T	M	O
								Max length						
V		P												
								Min length	D	E	L	T	M	O
								Max length						
V		P												
								Min length	D	E	L	T	M	O
								Max length						
V		P												
								Min length	D	E	L	T	M	O
								Max length						
V		P												
								Min length	D	E	L	T	M	O
								Max length						
V		P												
								Min length	D	E	L	T	M	O
								Max length						
V		P												
								Min length	D	E	L	T	M	O
								Max length						
V		P												

MKM: Rev/February 19, 2014

Attachment 6. IDEM OWQ Macroinvertebrate Header Form.

Office of Water Quality: Macroinvertebrate Header

L-Site #	Event ID	Stream Name	Location	County	Surveyor

Sample Date	Sample #	Macro#	# Containers

Macro Sample Type:
☐ Black Light ☐ Kick
☐ CPOM ☐ MHAB
☐ Hester-Dendy ☐ Qualitative

☐ Normal _____
☐ Duplicate _____
☐ Replicate _____

☐ Habitat Complete ☐ Sample Quality Rejected

Riparian Zone/Instream Features

Watershed Erosion:
☐ Heavy
☐ Moderate
☐ None

Watershed NPS Pollution:
☐ No Evidence
☐ Obvious Sources
☐ Some Potential Sources

Stream Depth Riffle (m):	Stream Depth Run (m):	Stream Depth Pool (m):	Distances Riffle-Riffle (m):	Distances Bend-Bend (m):

Stream Width (m):	High Water Mark (m):	Velocity (ft/s):

Stream Type:
☐ Cold
☐ Warm

Turbidity (Est):
☐ Clear ☐ Slightly Turbid
☐ Opaque ☐ Turbid

Salinity (mg/L): ☐

ORP (mV): ☐

☐ **Channelization** ☐ **Dam Present**

Predominant Surrounding Land Use: ☐ Forest ☐ Field/Pasture ☐ Agricultural ☐ Residential ☐ Commercial ☐ Industrial
Other ☐

Sediment

Sediment Odors: ☐ Normal ☐ Sewage ☐ Petroleum ☐ Chemical ☐ Anaerobic ☐ None Other ☐

Sediment Deposits: ☐ Sludge ☐ Sawdust ☐ Paper Fiber ☐ Sand ☐ Relic Shells Other ☐

Sediment Oils: ☐ Absent ☐ Moderate ☐ Profuse ☐ Slight

☐ Are the undersides of stones, which are not deeply embedded, black?

Substrate Components

(Note: Select from 0%, 10%, 20%, 30%, 40%, 50%, 60%, 70%, 80%, 90%, or 100% for each inorganic/ organic substrate component)

Inorganic Substrate Components (% Diameter)						
Bedrock	Boulder (>10 in)	Cobble (2.5-10 in)	Gravel (0.1-2.5 in)	Sand (gritty)	Silt	Clay (slick)

Organic Substrate Components (% Type)			
Detritus (sticks, wood)	Detritus (CPOM)	Muck/Mud (black, fine FPOM)	Marl(gray w/ shell fragments)

Water Quality

Water Odors: ☐ Normal ☐ Sewage ☐ Petroleum ☐ Chemical ☐ None Other ☐

Water Surface Oils: ☐ Slick ☐ Sheen ☐ Glob ☐ Flocks ☐ None

IDEM 03/14/13

Attachment 7. IDEM OWQ Biological Qualitative Habitat Evaluation Index (front).

OWQ Biological QHEI (Qualitative Habitat Evaluation Index)

IDEM

Sample # | bioSample # | Stream Name | Location

Surveyor | Sample Date | County | Macro Sample Type | ☐ Habitat Complete | QHEI Score:

1] *SUBSTRATE* Check ONLY Two predominant substrate TYPE BOXES; estimate % and check every type present

BEST TYPES

PREDOMINANT P/G R/R	PRESENT P/G R/R	TOTAL %
☐☐ BLDR/SLABS [10]	☐☐	____
☐☐ BOULDER [9]	☐☐	____
☐☐ COBBLE [8]	☐☐	____
☐☐ GRAVEL [7]	☐☐	____
☐☐ SAND [6]	☐☐	____
☐☐ BEDROCK [5]	☐☐	____

NUMBER OF BEST TYPES: ☐ 4 or more [2] ☐ 3 or less [0]

OTHER TYPES

PREDOMINANT P/G R/R	PRESENT P/G R/R	TOTAL %
☐☐ HARDPAN [4]	☐☐	____
☐☐ DETRITUS [3]	☐☐	____
☐☐ MUCK [2]	☐☐	____
☐☐ SILT [2]	☐☐	____
☐☐ ARTIFICIAL [0]	☐☐	____

(Score natural substrates; ignore sludge from point-sources)

Check ONE (Or 2 & average)

ORIGIN

☐ LIMESTONE [1]
☐ TILLS [1]
☐ WETLANDS [0]
☐ HARDPAN [0]
☐ SANDSTONE [0]
☐ RIP/RAP [0]
☐ LACUSTRINE [0]
☐ SHALE [-1]
☐ COAL FINES [-2]

QUALITY

SILT
☐ HEAVY [-2]
☐ MODERATE [-1]
☐ NORMAL [0]
☐ FREE [1]

EMBEDDEDNESS
☐ EXTENSIVE [-2]
☐ MODERATE [-1]
☐ NORMAL [0]
☐ NONE [1]

Substrate Maximum 20

Comments

2] *INSTREAM COVER* Indicate presence 0 to 3 and estimate percent: **0**–Absent; **1**–Very small amounts or if more common of marginal quality; **2**-Moderate amounts, but not of highest quality or in small amounts of highest quality; **3**-Highest quality in moderate or greater amounts (e.g., very large boulders in deep or fast water, large diameter log that is stable, well developed root wad in deep/fast water, or deep, well-defined, functional pools.)

% Amount		
___ ___ UNDERCUT BANKS [1]	___ ___ POOLS > 70cm [2]	___ ___ OXBOWS, BACKWATERS [1]
___ ___ OVERHANGING VEGETATION [1]	___ ___ ROOTWADS [1]	___ ___ AQUATIC MACROPHYTES [1]
___ ___ SHALLOWS (IN SLOW WATER) [1]	___ ___ BOULDERS [1]	___ ___ LOGS OR WOODY DEBRIS [1]
___ ___ ROOTMATS [1]		

AMOUNT
Check ONE (Or 2 & average)
☐ EXTENSIVE > 75% [11]
☐ MODERATE 25 - 75% [7]
☐ SPARSE 5 - < 25% [3]
☐ NEARLY ABSENT < 5% [1]

Cover Maximum 20

Comments

3] *CHANNEL MORPHOLOGY* Check ONE in each category (Or 2 & average)

SINUOSITY
☐ HIGH [4]
☐ MODERATE [3]
☐ LOW [2]
☐ NONE [1]

DEVELOPMENT
☐ EXCELLENT [7]
☐ GOOD [5]
☐ FAIR [3]
☐ POOR [1]

CHANNELIZATION
☐ NONE [6]
☐ RECOVERED [4]
☐ RECOVERING [3]
☐ RECENT OR NO RECOVERY [1]

STABILITY
☐ HIGH [3]
☐ MODERATE [2]
☐ LOW [1]

Channel Maximum 20

Comments

4] *BANK EROSION AND RIPARIAN ZONE* Check ONE in each category for EACH BANK (Or 2 per bank & average)

River right looking downstream

EROSION
L R
☐☐ NONE/LITTLE [3]
☐☐ MODERATE [2]
☐☐ HEAVY/SEVERE [1]

RIPARIAN WIDTH
L R
☐☐ WIDE > 50m [4]
☐☐ MODERATE 10-50m [3]
☐☐ NARROW 5-10m [2]
☐☐ VERY NARROW [1]
☐☐ NONE [0]

FLOOD PLAIN QUALITY
L R
☐☐ FOREST, SWAMP [3]
☐☐ SHRUB OR OLD FIELD [2]
☐☐ RESIDENTIAL, PARK, NEW FIELD [1]
☐☐ FENCED PASTURE [1]
☐☐ OPEN PASTURE, ROWCROP [0]

L R
☐☐ CONSERVATION TILLAGE [1]
☐☐ URBAN OR INDUSTRIAL [0]
☐☐ MINING/CONSTRUCTION [0]

Indicate predominant land use(s) past 100m riparian.

Riparian Maximum 10

Comments

5] *POOL/GLIDE AND RIFFLE/RUN QUALITY*

MAXIMUM DEPTH
Check ONE (ONLY!)
☐ > 1m [6]
☐ 0.7 - < 1m [4]
☐ 0.4 - < 0.7m [2]
☐ 0.2 - < 0.4m [1]
☐ < 0.2m [0] [metric = 0]

CHANNEL WIDTH
Check ONE (Or 2 & average)
☐ POOL WIDTH > RIFFLE WIDTH [2]
☐ POOL WIDTH = RIFFLE WIDTH [1]
☐ POOL WIDTH < RIFFLE WIDTH [0]

CURRENT VELOCITY
Check ALL that apply
☐ TORRENTIAL [-1]
☐ VERY FAST [1]
☐ FAST [1]
☐ MODERATE [1]
☐ SLOW [1]
☐ INTERSTITIAL [-1]
☐ INTERMITTENT [-2]
☐ EDDIES [1]
Indicate for reach – pools and riffles.

Recreation Potential
(Circle one and comment on back)
☐ Primary Contact
☐ Secondary Contact

Pool/ Current Maximum 12

Comments

Indicate for functional riffles; Best areas must be large enough to support a population of riffle-obligate species:

Check ONE (Or 2 & average) ☐ NO RIFFLE [metric = 0]

RIFFLE DEPTH
☐ BEST AREAS > 10cm [2]
☐ BEST AREAS 5 - 10cm [1]
☐ BEST AREAS < 5cm [metric = 0]

RUN DEPTH
☐ MAXIMUM > 50cm [2]
☐ MAXIMUM < 50cm [1]

RIFFLE/RUN SUBSTRATE
☐ STABLE (e.g., Cobble, Boulder) [2]
☐ MOD. STABLE (e.g., Large Gravel) [1]
☐ UNSTABLE (e.g., Fine Gravel, Sand) [0]

RIFFLE/RUN EMBEDDEDNESS
☐ NONE [2]
☐ LOW [1]
☐ MODERATE [0]
☐ EXTENSIVE [-1]

Riffle/ Run Maximum 8

Comments

6] *GRADIENT* (ft/mi)

DRAINAGE AREA (mi²)

☐ VERY LOW - LOW [2 – 4]
☐ MODERATE [6 - 10]
☐ HIGH - VERY HIGH [10 - 6]

%POOL: ____ %GLIDE: ____
%RUN: ____ %RIFFLE: ____

Gradient Maximum 10

IDEM 11/15/12

Attachment 7 (continued). IDEM OWQ Biological QHEI (back).

OWQ Biological QHEI (Qualitative Habitat Evaluation Index)

COMMENT _____

A-CANOPY
- ☐ >85% - Open
- ☐ 55% - <85%
- ☐ 30% - <55%
- ☐ 10% - <30%
- ☐ <10% - Closed

B-AESTHETICS
- ☐ Nuisance algae
- ☐ Invasive macrophytes
- ☐ Excess turbidity
- ☐ Discoloration
- ☐ Foam/Scum
- ☐ Oil sheen
- ☐ Trash/Litter
- ☐ Nuisance odor
- ☐ Sludge deposits
- ☐ CSOs/SSOs/Outfalls

C-RECREATION

Area Depth
Pool ☐ > 100 ft² ☐ > 3 ft

D-MAINTENANCE
- ☐ Public ☐ Private
- ☐ Active ☐ Historic
- Succession: ☐ Young ☐ Old
- ☐ Spray ☐ Islands ☐ Scoured
- Snag: ☐ Removed ☐ Modified
- Leveed: ☐ One sided ☐ Both banks
- ☐ Relocated ☐ Cutoffs
- Bedload: ☐ Moving ☐ Stable
- ☐ Armoured ☐ Slumps
- ☐ Impounded ☐ Desiccated
- ☐ Flood control ☐ Drainage

E-ISSUES
- ☐ WWTP ☐ CSO ☐ NPDES
- ☐ Industry ☐ Urban
- ☐ Hardened ☐ Dirt & Grime
- ☐ Contaminated ☐ Landfill
- BMPs: ☐ Construction ☐ Sediment
- ☐ Logging ☐ Irrigation ☐ Cooling
- Erosion: ☐ Bank ☐ Surface
- ☐ False bank ☐ Manure ☐ Lagoon
- ☐ Wash H_2O ☐ Tile ☐ H_2O Table
- Mine: ☐ Acid ☐ Quarry
- Flow: ☐ Natural ☐ Stagnant
- ☐ Wetland ☐ Park ☐ Golf
- ☐ Lawn ☐ Home
- ☐ Atmospheric deposition
- ☐ Agriculture ☐ Livestock

Looking upstream (> 10m, 3 readings; ≤ 10m, 1 reading in middle); Round to the nearest whole percent

	Right	Middle	Left	Total Average
% open	___ %	___ %	___ %	___ %

Stream Drawing:

IDEM 11/15/12

Attachment 8. IDEM OWQ Chain of Custody Form.

IDEM Indiana Department of Environmental Management
OWQ Chain of Custody Form

Project: ______________________
Team: ______________________
OWQ Analysis Set: ______________________

I Certify that the sample(s) listed below was/were collected by me, or in my presence.

Date: ______________________

Signature: ____

Section: ______________________

Lab Assigned Number	IDEM Control Number	Sample Type	ID	1000 ml P, N. M.	1000 ml G, N. M.	40 ml VIAL	120 ml G (Bact)	500 ml P, N. M.	250 ml P, N. M.			Date And Time Collected	
												Date	Time

P = Plastic G = Glass N. M. = Narrow Mouth Bact = Bacteriological Only Should samples be iced? Y N

Carriers

I certify that I have received the above sample(s).

Signature	Date	Time	Seals Intact		Comments
Relinquished By:			Y	N	
Received By:					
Relinquished By:			Y	N	
Received By:					
Relinquished By:			Y	N	
Received By:					

Lab Custodian

I certify that I have received the above sample(s), which has/have been recorded in the official record book. The same sample(s) will be in the custody of competent laboratory personnel at all times, or locked in a secured area.

Signature: ______________________________ Date: ____________ Time: ____________

Lab: ______________________ Address: ______________________________

04/17/2003 12:31:20 PM 04/17/2003 12:31:20 PM Fixed Station Chain of Custody Datasheet, Page 1 of 1

Attachment 9. 2016 Reference Sites Water Sample Analysis Request Form.

Indiana Department of Environmental Management
Office of Water Quality
Watershed Planning and Assessment Branch
www.idem.IN.gov

Water Sample Analysis Request

Project Name: 2016 Reference Sites **Composite ☐ Grab ☒**

OWQ Sample Set	16SPW	IDEM Sample Nos.	
Crew Chief		Lab Sample Nos.	
Collection Date	Apr. - Oct.	Lab Delivery Date	

Anions and Physical Parameters

Parameter	Test Method	Total	Dissolved
Alkalinity	310.2	☒ **	☐
Total Solids	SM2540B	☒ **	
Suspended Solids	SM2540D	☒ **	
Dissolved Solids	SM2540C		☒ **
Sulfate	300.0	☐ **	☒ **
Chloride	300.0	☐ **	☒
Hardness (Calculated)	SM-2340B	☒ **	☐
Fluoride	SM4500-F-C	☐ **	☐

Priority Pollutant Metals Water Parameters

Parameter	Test Method	Total	Dissolved
Antimony	200.8	☒	☒
Arsenic	200.8	☒	☒
Beryllium	200.8	☐	☐
Cadmium	200.8	☒	☒
Chromium	200.7	☒	☒
Copper	200.8	☒	☒
Lead	200.8	☒	☒
Mercury, Low Level	1631, Rev E.	☐	☐
Nickel	200.8	☒	☒
Selenium	200.8	☒	☒
Silver	200.8	☒	☒
Thallium	200.8	☐	☐
Zinc	200.7	☒	☒

Cations and Secondary Metals Parameters

Parameter	Test Method	Total	Dissolved
Aluminum	**200.7**, 200.8	☒	☒
Barium	200.8	☐	☐
Boron	200.8	☐	☐
Calcium	**200.7**, 200.8	☒ ***	☐
Cobalt	200.8	☐	☐
Iron	200.7	☐	☐
Magnesium	**200.7**, 200.8	☒ ***	☐
Manganese	200.8	☐	☐
Sodium	200.7	☐	☐
Silica, Total Reactive	200.7	☐	☐
Strontium	200.8	☐	☐

Send reports (Fed. Ex. or UPS) to:
Tim Bowren - IDEM
STE 100
2525 North Shadeland Ave.
Indianapolis, IN 46219

Deliver reports to:
Tim Bowren – IDEM
STE 100
2525 North Shadeland Ave.
Indianapolis, IN 46219

Organic Water Parameters

Parameter	Test Method	Total
Priority Pollutants: Oranochlorine Pesticides and PCBs	608	☐
Priority Pollutants: VOCs - Purgeable Organics	624	☐
Priority Pollutants: Base/Neutral Extractables	625	☐
Priority Pollutants: Acid Extractables	625	☐
Phenolics, 4AAP	420.4	☐
Oil and Grease, Total	1664A	☐

Nutrient & Organic Water Chemistry Parameters

Parameter	Test Method	Total	Dissolved
Ammonia Nitrogen	SM4500NH3-G	☒	☐
$CBOD_5$	SM5210B	☐	
Total Kjeldahl Nitrogen (TKN)	SM4500N(Org)	☒	☐
Nitrate + Nitrite	353.2	☒	☐
Total Phosphorus	365.1	☒	☐
TOC	SM 5310C	☒	☐
COD	410.4	☒	☐
Cyanide (Total)	335.4	☐	☐
Cyanide (Free)	SM4500CN-I	☐ *	☐
Cyanide (Amenable)	SM4500CN-G	☐ *	☐
Sulfide, Total	376.2	☐	☐

RFP 12-48 Contract Number:	A305-3-1 (Pace-Indy) PO # 0014561536 (Pace-Indy)

30 day reporting time required.

Notes:
** **= DO NOT RUN PARAMETER IF SAMPLE IDENTIFIED AS A BLANK ON THE CHAIN OF CUSTODY**
* **= RUN ONLY IF TOTAL CYANIDE IS DETECTED**
*** *= Report Calcium, Magnesium as Total Hardness components*

Testing Laboratory: Pace Analytical Services, Inc.
Attn: Sue Brotherton
Phone: 317-228-3136 7726 Moller Road
Indianapolis, IN 46268

Attachment 10. IDEM Biological Samples Field Chain of Custody Form.

IDEM

INDIANA DEPARTMENT OF
ENVIRONMENTAL MANAGEMENT
OFFICE OF WATER QUALITY
BIOLOGICAL STUDIES SECTION
BIOLOGICAL SAMPLES FIELD CHAIN OF CUSTODY

I certify that the sample(s) listed below was/were collected by me or in my presence.

Signature:

Date: ____/____/____
Sample Type:
(ie. fish, macro, algae)

EVENT ID (YY____)/ MACRO # (9 DIGIT)	IDEM SAMPLE # (AB_____)	2000 mL Nalgene	250mL Nalgene	125mL Glass	Type (AD or AS)	Volume (mL)	COLLECTED		PLACED IN STORAGE		Storage Room #	Check line for sample present and accounted for! One check per bottle.
							Date (mm/dd/yyyy)	Time (24 hr)	Date (mm/dd/yyyy)	Time (24 hr)		
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SIGNATURE	DATE AND TIME	Comments:
RELINQUISHED BY:	/ /	
RECEIVED BY:	:	
RELINQUISHED BY:	/ /	
RECEIVED BY:	:	

Lab Custodian

I certify that I received the above sample(s) and is/are recorded in the official record book. The same samples will be in custody of competent laboratory personnel at all times or locked in a secure area.

Signature: ____ Date: / / Time: :

Lab: Indiana Department of Environmental Management	Address: 2525 N. Shadeland Ave., Indianapolis, IN 46219

Appendix 1. List of IDEM Documents and SOPs used in the development of the 2016 Reference Site Monitoring Program Sampling and Analysis Work Plan.

Indiana Department of Environmental Management (IDEM). 1992, revision 1. Section 2, Biological Studies Section Hazards Communications Manual (List of Contents). Pages 74 – 91 *in* IDEM. 1992. Biological Studies Section Standard Operating Procedures Manual (SOP). Biological Studies Section, Surveillance and Standards Branch, Office of Water Management (OWM), IDEM, Indianapolis, Indiana.

IDEM. 1992, revision 1. Section 3, Quality Assurance Project Plan, Development of Biological Criteria (Fish) for the Ecoregions of Indiana. Biological Studies Section, Surveillance and Standards Branch, OWM, IDEM, Indianapolis, Indiana.

IDEM. 1992, revision 1. Section 4, Standard Operating Procedures for Fish Collections, Use of Seines, Electrofishers, and Sample Processing. Pages 496 – 534 *in* IDEM. 1992. Biological Studies Section Standard Operating Procedures Manual (SOP). Biological Studies Section, Surveillance and Standards Branch, OWM, IDEM, Indianapolis, Indiana.

IDEM. 1992, revision 1. Section 5, Standard Operating Procedures for Conducting Rapid Assessment of Ambient Water Quality Using Fish (RBP-V). Pages 535 – 663 *in* IDEM. 1992. Biological Studies Section Standard Operating Procedures Manual (SOP). Biological Studies Section, Surveillance and Standards Branch, OWM, IDEM, Indianapolis, Indiana

IDEM. 1992, revision 1. Section 11, Standard Operating Procedures-Appendices of Operational Equipment Manuals and Procedures. Page 1386 – 3313 *in* IDEM. 1992. Biological Studies Section Standard Operating Procedures Manual (SOP). Biological Studies Section, Surveillance and Standards Branch, OWM, IDEM, Indianapolis, Indiana.

IDEM. 1997. Water Quality Surveys Section Laboratory and Field Hazard Communication Plan Supplement. IDEM 032/02/018/1998, Revised October 1998. Assessment Branch, IDEM, Indianapolis, Indiana.

IDEM. 2002. Water Quality Surveys Section Field Procedure Manual, Assessment Branch, IDEM, Indianapolis, Indiana. Located at https://extranet.idem.in.gov/standards/docs/quality_improvement/qapps/owq_surveys_section_field_manual.pdf

IDEM. 2004. Quality Assurance Project Plan (QAPP) for Indiana Surface Water Quality Monitoring and Total Maximum Daily Load (TMDL) Program, (Rev. 3, Oct. 2004). Located at the IDEM OWQ WAPB offices (100 North Senate Avenue, Indianapolis, IN 46204-2251).

IDEM. 2008. IDEM Personal Protective Equipment Policy, revised May 1 2008. A-059-OEA-08-P-R0. IDEM, Indianapolis, Indiana.

IDEM. 2010. Multi-habitat (MHAB) Macroinvertebrate Collection Procedure Technical Standard Operating Procedure. S-001-OWQ-W-BS-10-T-R0. Office of Water Quality, Watershed Planning and Assessment Branch, Biological Studies Section. Indianapolis, Indiana. Located at https://extranet.idem.in.gov/standards/docs/sops/owq/S-001-OWQ-W-BS-10-S-R0.pdf

IDEM. 2010. IDEM Health and Safety Training Policy, revised October 1 2010. A-030-OEA-10-P-R2. IDEM, Indianapolis, Indiana.

IDEM. 2010. IDEM Injury and Illness Resulting from Occupational Exposure Policy, revised October 1 2010. A-034-OEA-10-P-R2. IDEM, Indianapolis, Indiana.

IDEM. 2012a. Request for Proposals 12-48, Solicitation for Analyses. IDEM. Indiana Department of Administration. Indianapolis, Indiana.

IDEM. 2012b.IDEM Agency Wide Quality Management Plan. IDEM, Indiana government Center North, 100 N. Senate Ave., Indianapolis, Indiana, 46204. Available at http://www.in.gov/idem/files/idem_qmp_2012.pdf

IDEM. 2014. Indiana Integrated Water Monitoring and Assessment Report 2014. Edited by Jody Arthur. Office of Water Quality, IDEM, Indianapolis, Indiana.

IDEM. 2014. Indiana's 2014 Consolidated Assessment and Listing Methodology (CALM) Revised. Edited by Jody Arthur. Office of Water Quality, IDEM, Indianapolis, Indiana.

IDEM. 2015a. Processing and Identification of Diatom Samples Technical Standard Operating Procedure. B-002-OWQ-WAP-TGM-15-T-R0. Office of Water Quality, Watershed Assessment and Planning Branch. Indianapolis, Indiana. Located at https://extranet.idem.in.gov/standards/docs/sops/owq/B-002-OWQ-WAP-TGM-15-T-R0.pdf

IDEM. 2015b. Global Positioning System (GPS) Data Creation Technical Standard Operating Procedure. B-001-OWQ-WAP-XXX-15-T-R0. Office of Water Quality, Watershed Assessment and Planning Branch. Indianapolis, Indiana. Located at https://extranet.idem.in.gov/standards/docs/sops/owq/B-001-OWQ-WAP-XXX-15-T-R0.pdf

IDEM. 2016. Phytoplankton and Periphyton Field Collection Procedures. B-004-OWQ-WAP-XX-16-T-R0. Office of Water Quality, Watershed Assessment and Planning Branch. Indianapolis, Indiana. Located at https://extranet.idem.in.gov/standards/docs/sops/owq/B-004-OWQ-WAP-XX-16-T-R0.pdf

Appendix 2. IDEM Fish Community Assessments for Aquatic Life Use.

IDEM collects fish along with other data (chemical parameters, nutrients, macroinvertebrate, and habitat) to monitor the health of streams and rivers in Indiana. There are many advantages of using fish for monitoring stream health:

- Many fish have life spans of greater than three years, allowing detection of degradation in habitat or water chemistry over time which will alter the expected fish community structure.
- The knowledge of fish life history, feeding, and reproductive behavior is well known and can be used to detect changes in water chemistry or habitat alterations.
- Identification of fish species can usually be made in the field so that fish are returned to the stream and time utilized for laboratory identifications kept minimal.

The Indiana Administrative Code [327 IAC 2-1-3(a)(2); 327 IAC 2-1.5-5(a)(2)] has narrative biological criteria that states "all waters, except those designated as limited use, will be capable of supporting a well-balanced, warm water aquatic community." The water quality standard definition of a "well-balanced aquatic community" is "an aquatic community that is diverse in species composition, contains several different trophic levels, and is not composed mainly of pollution tolerant species" [327 IAC 2-1-9(59)]. To measure whether or not the fish community meets this definition, IDEM uses an Index of Biotic Integrity (IBI), which is composed of 12 fish community characteristics chosen based on what part of the state you are sampling (ecoregion) and the size of stream (drainage area). The 12 different characteristics can score a 0, 1, 3, or 5, each of which represents a deviation from expected fish community structure (i.e. 5 = no deviation from expectations, 1 = severe deviation from expected fish community structure). The total score can range from 0 (no fish) to 60 (excellent, comparable to "least impacted" conditions). Indiana expects streams to score at least 36 out of 60 to meet aquatic life use water quality standards. The chart below, modified from a table developed by Karr et al. 1986, uses total IBI score, integrity class and attributes to define the fish community characteristics in Indiana streams and rivers.

Total IBI Score	Integrity Class	Attributes
53-60	Excellent	Comparable to "least impacted" conditions, exceptional assemblage of species.
45-52	Good	Decreased species richness (intolerant species in particular), sensitive species present.
36-44	Fair	Intolerant and sensitive species absent, skewed trophic structure.
23-35	Poor	Top carnivores and many expected species absent or rare, omnivores and tolerant species dominant.
12-22	Very Poor	Few species and individuals present, tolerant species dominant, diseased fish frequent.
<12	No Fish	No fish captured during sampling.

Karr, J.R., K.D. Fausch, P.L. Angermeier, P.R. Yant, and I.J Schlosser. 1986. Assessing biological integrity in running waters: a method and its rationale. Illinois Natural History Survey Special Publication 5. 28 p.

Some examples of metrics and fish specimens for the Index of Biotic Integrity (IBI) looking at species composition, trophic levels, and tolerance to water pollution or habitat disturbance.

1. *Number of Species* (generally more species = better quality stream)

2. *Number of Darter, Madtom, Sculpin Species* (species require high dissolved oxygen and clean rocky substrates so higher number = better quality stream)
   - Examples: rainbow darter, brindled madtom, mottled sculpin

   % Large River Individuals (species require habitats typical in great rivers in terms of bottom substrates, current velocity, backwater areas, etc., so higher percentage = better quality river)
   - Examples: chestnut lamprey, channel catfish, bullhead minnow, silver chub

3. *% Headwater Individuals* (species in small streams occupying permanent habitat with low environmental stress so greater percentage = better quality stream)
   - Examples: western blacknose dace, southern redbelly dace, fantail darter

   Number of Sunfish or Centrarchidae Species (species occupy pools which act as "sinks" for potential pollutants and silt so fewer number of these species = low quality stream)
   - Examples: rock bass, bluegill, largemouth bass

4. *Number of Sucker or Round Body Sucker Species* (species do not tolerate habitat and water quality degradation so more = better quality stream)
   - Examples: black redhorse, northern hog sucker

   Number of Minnow Species (generally more minnow species = better quality stream)
   - Examples: spotfin shiner, silverjaw minnow, hornyhead chub

5. *Number of Sensitive Species* (species sensitive to pollution so more species = better quality stream)
   - Examples: greenside darter, smallmouth bass, longear sunfish

6. *% Tolerant Individuals* (species tolerant to pollution so greater percentage = low quality stream)
   - Examples: yellow bullhead, green sunfish, central mudminnow

7. *% Omnivore/Detritivore Individuals* (species that consume at least 25% plant and 25% animal material which makes them opportunistic feeders when other food sources are scarce; thus, greater percentage = lower quality stream)
   - Examples: bluntnose minnow, white sucker, gizzard shad

8. *% Insectivore/Invertivore Individuals* (species whose diet is mainly benthic insects so the metric is a reflection of the food source; thus, lower percentage = lower quality stream)
   - Examples: blackstripe topminnow, emerald shiner, logperch

9. *% Carnivore Individuals* (species whose diet is carnivorous and also reflects the availability of the food source; too high or too low percentage of carnivores = lower quality stream and imbalance of trophic levels)
   - Examples: spotted bass, grass pickerel

   % Pioneer Individuals (species that are first to colonize a stream after environmental disturbance so higher percentage of pioneer individuals = lower quality stream)
   - Examples: creek chub, central stoneroller, johnny darter

10. *Number of Individuals* (generally more individuals = better quality stream)

11. *% Simple Lithophilic Individuals* (species that require clean gravel or cobble for successful reproduction since they simply broadcast their eggs on the substrate, fertilize, and provide no parental care; thus, heavy siltation or environmental disturbance will result in a lower percentage of simple lithophilic species = lower quality stream)
    - Examples: bigeye chub, striped shiner, orangethroat darter

12. *% Individuals with Deformities, Eroded Fins, Lesions, and Tumors (DELT's)* (diseased individuals with external anomalies as a result of bacterial, fungal, viral, and parasitic infections, chemical pollutants, overcrowding, improper diet, and other environmental degradation. Percentages should be absent or very low naturally so higher percentage = low quality stream)
    - Examples: deformed blackstripe topminnow, creek chub with tumors

Appendix 3. Calculating IDEM Macroinvertebrate Index of Biotic Integrity (mIBI).

The purpose of this document is to describe the laboratory processing and data analysis procedures used by the Indiana Department of Environmental Management (IDEM) to calculate the macroinvertebrate Index of Biotic Integrity (mIBI). Standard Operating Procedures (SOPs) are being developed to describe these processes but it may be some time before they are finalized.

An SOP describing the methods used by IDEM to collect macroinvertebrate samples with a multi-habitat (MHAB) sampling method was recently completed (available at http://monitoringprotocols.pbworks.com/f/S-001-OWQ-W-BS-10-S-R0.pdf). The index period for collection of macroinvertebrate samples with the MHAB sampling method is July 15 to October 30. The entire sample is processed in the laboratory as subsampling has already been performed in the field. All macroinvertebrate individuals are counted with the exception of empty snail and clam shells, micro-crustaceans (Ostracoda, Branchiopoda, Copepoda), larval and pupal insect exuviae, and terrestrial insects (including the terrestrial adults of aquatic insect larvae); invertebrate specimens missing their head are also excluded.

The level of taxonomic resolution used in the identification of macroinvertebrates may depend in large part on the condition (instar and physical condition) of the specimens and the availability of taxonomic resources that are comprehensive and appropriate for Indiana's fauna. Specimens are generally identified to the “lowest practical" taxonomic level. Oligochaeta (aquatic worms, Hirudinea and Branchiobdellida), Planaria and Acari are only identified to family or a higher level; freshwater snails and clams are identified to genus; freshwater crustacea are identified to genus (Amphipoda and Isopoda) or species (Decapoda); aquatic insects are identified to family (Collembola and several Dipteran families) or genus and species (all other insects). The following table lists insect genera that are often identified to species (and may contain multiple species in a sample) and taxonomic resources commonly used by IDEM biologists for their identification (full citations for these resources are listed in the Taxonomic References at the end of this document.

Ephemeroptera:
Baetidae: *Baetis* (separate *B. intercalaris* and *B. flavistriga* with Moriharra and McCafferty 1979, leave everything else at *Baetis)*
Caenidae: *Caenis:* Provonsha 1990
Heptageniidae: *Mccaffertium* (formerly *Stenonema* subgenus *Mccaffertium*): Bednarik and McCafferty 1979
Odonata:
Gomphidae: *Dromogomphus*: Westfall and Tennessen 1979
Coenagrionidae: *Argia* and *Enallagma*: Westfall and May 1996
Hemiptera:
Corixidae: *Trichocorixa* and *Palmacorixa*: Hungerford 1948, Hilsenhoff 1984
Megaloptera:
Corydalidae: *Chauliodes* and *Nigronia*: Rasmussen and Pescador 2002
Coleoptera:
Haliplidae: *Peltodytes*: Brigham 1996
Dytiscidae: *Neoporus*, *Heterosternuta*, *Laccophilus*, *Coptotomus*: Larson et al. 2000.
Hydrophilidae: *Tropisternus*, *Berosus*, *Enochrus*: Hilsenhoff 1995A and 1995B.
Elmidae: *Stenelmis*, *Dubiraphia*, *Optioservus*: Hilsenhoff and Schmude, Hilsenhoff 1982
Trichoptera:
Philopotamidae: *Chimarra*: Hilsenhoff 1982
Leptoceridae: *Nectopsyche*: Glover and Floyd 2004
Hydropsychidae: *Hydropsyche*: Schuster and Etnier 1978
Diptera:
Chironomidae: *Ablabesmyia*: Roback 1985 (sub-genus/ species group)
Polypedilum: Maschwitz and Cook 2000 (sub-genus/ species group)
Cricotopus/Orthocladius: Merritt et al 2007 (sub-genus/ species group)

After all organisms in the sample have been identified to the lowest practical taxon, those taxa are then associated with their corresponding tolerance, functional feeding group and habit values (found in the spreadsheet "Indiana Macroinvertebrate Attributes"). Organisms without a tolerance value, functional feeding group, or habit are not included in the calculations for those specific metrics (this may become more evident while looking at the metric example provided). For taxa metrics, all of the taxa listed for a specific group (EPT, Diptera) are counted, regardless of level of identification (i.e.,. if there were 4 taxa under the Chironomidae family (1 family level ID, 1 *Cricotopus* genus level ID, and 2 distinct species level IDs under the *Cricotopus* genus) this would be considered 4 taxa).

The metrics are then calculated as follows:
1 - Total Number of Taxa: Numerical count of all identified taxa in the sample
2 - Total Number of Individuals: Numerical count of the number of individual specimens in the sample
3 - Total Number of EPT Taxa: Numerical count of all Ephemeroptera, Plecoptera and Trichoptera taxa in the sample
4 - Total Number of Diptera Taxa: Numerical count of all Diptera taxa in the sample
5 - % Orthocladiinae + Tanytarsini of Chironomidae: Number of individuals in the chironomid subfamily Orthocladiinae and tribe Tanytarsini divided by the total number of Chironomidae in the sample
6 - % Non-insect (minus crayfish): Number of individuals, except for crayfish, that are not in the Class Insecta (Isopoda, Amphipoda, Acari, snails, freshwater clams, Oligochaeta, Nematoda, Nematomorpha) divided by the total number of individuals in the sample
7 - % Intolerant: Number of individuals with a tolerance value of 0-3 divided by the total number of individuals in the sample
8 - % Tolerant: Number of individuals with a tolerance value of 8-10 divided by the total number of individuals in the sample
9 - % Predators: Number of individuals with a functional feeding group designation of "Predator" divided by the total number of individuals in the sample
10 - % Shredders + Scrapers: Combined number of individuals in the functional feeding groups "Shredder" and "Scraper" divided by the total number of individuals in the sample
11 - % Collector-Filterers: Number of individuals in the functional feeding group "Collector-Filterer" divided by the total number of individuals in the sample
12 - % Sprawlers: Number of individuals with a habit specificity of "Sprawler" divided by the total number of individuals in the sample

These metric values are then scored as a 1, 3 or 5 according to the criteria in the following table:

Metric	1	3	5
Number of Taxa	< 21	≥ 21 and <41	≥ 41
Number of Individuals	< 129	≥ 129 and < 258	≥ 258
Number of EPT Taxa			
Drainage Area: < 5 mi^2	< 2	≥ 2 and < 4	≥ 4
Drainage Area: ≥ 5 and < 50 mi^2	< 4	≥ 4 and < 8	≥ 8
Drainage Area: ≥ 50 mi^2	< 6	≥ 6 and < 12	≥ 12
% Orthocladiinae + Tanytarsini of Chironomidae	≥ 47	≥ 24 and < 47	< 24
% Non-insects Minus Crayfish	≥ 35	≥ 18 and < 35	< 18
Number of Diptera Taxa	< 7	≥ 7 and < 14	≥ 14
% Intolerant	< 15.9	≥ 15.9 and < 31.8	≥ 31.8
% Tolerant	≥ 25.3	≥ 12.6 and < 25.3	< 12.6
% Predators	< 18	≥ 18 and < 36	≥ 36
% Shredders + Scrapers	< 10	≥ 10 and < 20	≥ 20
% Collector-Filterers	≥ 20	≥ 10 and < 20	< 10
% Sprawlers	< 3	≥ 3 and < 6	≥ 6

Most scoring classifications are the same regardless of stream drainage area; the exception is the "Number of EPT Taxa" metric which increases with increasing drainage area. After all metrics have been scored, the individual metric scores are summed and the total is the mIBI score for that particular site. Scores less than 36 are considered impaired while those greater than or equal to 36 are unimpaired.

Example of Derivation of Metric Scores for the Macroinvertebrate Index of Biotic Integrity

TAXA NAME	FEED GRP	TOL	HAB/BHV	# OF IND
Heptagenia	SC	3		1
Leucrocuta	SC	2	cn	1
Acerpenna pygmaea	OM	2	sw	1
Baetis flavistriga	GC	3	sw	1
Callibaetis	GC	6	sw	1
Ephemera simulans				1
Ischnura verticalis	PR			1
Berosus peregrinus	SH	6	sw	1
Dubiraphia	GC	5	cn	1
Macronychus glabratus	OM	3	cn	1
Ceratopsyche bronta		5		1
Pycnopsyche	SH	3	sp	1
Chrysops	GC	5		1
Procladius	PR	7	sp	1
Paraphaenocladius	GC		sp	1
Lirceus	GC	8	cr	1
Ferrissia rivularis	SC	6		1
Physella	SC	8		1
Corbicula fluminea	FC	6		1
NAIDIDAE	GC	8		1
Acariformes		4		1
Maccaffertium pulchellum	SC	2		2
Tricorythodes	GC	3	sw	2
Boyeria vinosa	PR	4	cb	2
Rheumatobates	PR		sk	2
Trepobates	PR			2
Stenelmis	SC	5	cn	2
Polypedilum flavum				2
Stictochironomus	OM	4	bu	2
Caenis latipennis	GC			3
Palmacorixa nana	PI	4	sw	3
Cheumatopsyche	FC	3	cn	3
Orconectes	GC	4		3
Hetaerina americana	PR			4
Ancyronyx variegatus	OM	4		5
Baetis intercalaris	OM	3	sw	6
Peltodytes duodecimpunctata				6
Trepobates inermis				7
Dubiraphia minima				7
Hyalella azteca	GC	8	cr	9
Polypedilum illinoense		7		16
Stenelmis sexlineata				18
Grand Total				127

Metrics	Metric Values	Metric Scores
Total Number of Taxa	42	**3**
Total Abundance of Individuals	127	**1**
Number of EPT Taxa	13	**5**
% Orthocladinae + Tanytarsinii of Chironomidae	4.55	**5**
% Non-Insects - Crayfish	11.81	**5**
Number of Diptera Taxa	6	**1**
% Intolerant Taxa (Score 0 - 3)	14.96	**1**
% Tolerant Taxa (Score 8 - 10)	9.45	**5**
% Predators	9.45	**1**
% Shredders + Scrapers	7.87	**1**
% Collector-Filterers	3.15	**5**
% Sprawlers	2.36	**1**
MIBI Score		**34**

Taxonomic References

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If you have further questions regarding the IDEM mIBI please contact:

Paul D. McMurray, Jr.
Environmental Manager
Indiana Department of Environmental Management
Office of Water Quality, Watershed Assessment and Planning Branch
Probabilistic Monitoring Section
100 N. Senate Ave.
MC 65-40-2 Shadeland
Indianapolis, IN 46204-2251
317-308-3210
pmcmurra@idem.in.gov

Todd E. Davis
Environmental Manager
Indiana Department of Environmental Management
Office of Water Quality, Watershed Assessment and Planning Branch
Probabilistic Monitoring Section
100 N. Senate Ave.
MC 65-40-2 Shadeland
Indianapolis, IN 46204-2251
317-308-3188
tdavis@idem.in.gov