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BIOLOGICAL EVALUATION OF NON-WADEABLE RIVERS IN MICHIGAN

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KELLY JAMES WESSELL

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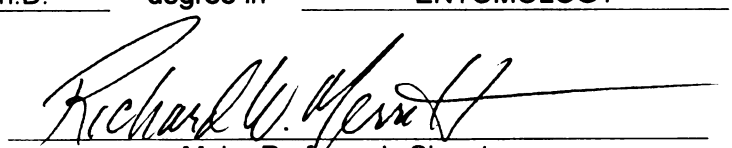

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BIOLOGICAL EVALUATION OF NON-WADEABLE RIVERS IN MICHIGAN

By

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ABSTRACT

BIOLOGICAL EVALUATION OF NON-WADEABLE RIVERS IN MICHIGAN

By

Kelly James Wessell

Compared to smaller, wadeable streams, non-wadeable rivers are relatively understudied. Currently, protocols exist in most states, including Michigan, to evaluate the ecological condition of wadeable streams, but none such protocols exist for larger, non-wadeable rivers. The goal of this research was to establish sampling protocols and develop a multimetric index of biological integrity for Michigan's non-wadeable rivers. I sampled 28 unique non-wadeable river reaches in Michigan that encompassed a wide range of human impacts and ecological conditions. In each reach, I took physical, chemical, and macroinvertebrate samples. I found that sample reaches had unique physical, chemical, and biological characteristics that allowed the evaluation of ecological health at the reach scale. Using several techniques to eliminate redundancy among metrics and identify those biological attributes that accounted for the most among-reach variation in macroinvertebrate communities, I developed a useful protocol that will allow the rapid bioassessment of non-wadeable rivers in Michigan. When used together with the Habitat Index, the NW-IBI will allow the objective evaluation of non-wadeable rivers that may be applicable to other regions.

This dissertation is dedicated to my grandparents, Helen and James Hines, Tyrus Wessell, Sr., and Alice Bennett. If only everyone could be so lucky....

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INTRODUCTION AND LITERATURE REVIEW

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INTRODUCTION AND LITERATURE REVIEW

INTRODUCTION

Large river ecosystems have long been associated with the development of human civilization. They provide water for irrigation of agricultural land, help in replenishing nutrients on their floodplains, and carry away waste generated by inhabitants of their watersheds. Humans have historically settled in areas within close proximity to rivers for the above reasons, and have invariably modified and impacted these systems on which they rely so heavily. As the ultimate sink for all upstream and watershed-wide processes, large rivers have been subjected to many different types of impacts from their human inhabitants including: 1) nutrient enrichment; 2) other non-point source pollution like pesticide inputs and sedimentation from intensive agriculture; 3) channel modification for navigation and drainage purposes; and 4) industrial pollution from point-source discharge of waste. Since the Clean Water Act, state and federal agencies in the United States have established various procedures to protect and evaluate lotic ecosystem integrity. With the application of scientific theory to set expectations for river health and monitoring of current conditions and trends, our riverine systems are slowly improving, but this progress has been extremely biased toward smaller, wadeable streams.

LARGE RIVER ECOLOGY

Much of the science of stream ecology has been developed on small streams. This is due to several factors: Smaller streams are easier environments to sample, because there is no need for a boat or access sites. Also, many stream ecologists focus on

working in pristine streams, free of anthropogenic influences. One is hard-pressed to find a large river that is not affected by humans, whether these effects are in terms of watershed land use, impoundments, or channel modification. Despite large rivers being relatively understudied compared to small streams, there have been several important theories developed in the last 50 years that have advanced our understanding of how large rivers are structured and how they function.

River Continuum Concept

In their classic paper, Vannote et al. (1980) synthesized what was known about the structure and function of lotic ecosystems by describing an orderly shift in energy sources and consumer groups along a predictable gradient of physical factors from headwaters to mouth. Specifically, the river continuum concept (RCC) assumes that energy for biological production comes from three sources: local inputs of organic matter form riparian sources (allochthonous inputs), primary production from within the stream (autochthonous inputs), and transport of organic matter from upstream. The relative importance of each energy source varies along the river continuum, and is predicable based on a dynamic equilibrium with the physical environment (Johnson et al. 1995a;Vannote et al. 1980).

Small, low order streams have a relatively constant environment due to continuous groundwater inputs and small watershed area. Within a forest, the canopy reduces the amount of light the stream receives, thus limiting photosynthesis. Because of this, the RCC predicts that the dominant energy source for low order (orders 1-3) streams will be coarse particulate organic matter (CPOM) derived from terrestrial leaf litter that

falls into the stream. As a result, the primary macroinvertebrate groups in these small streams are shredders who feed on the CPOM and collectors who filter out fine particles from the water column.

The physical processes of mid-sized rivers (order 4-6) are more variable, exhibiting the largest range of temperatures and hydraulic conditions. The forest canopy opens, allowing light available for autochthonous production, but reducing leaf litter inputs into the stream. This shift in energy source results in a shift in the dominant invertebrate functional feeding groups present. Here, scrapers and collectors will dominate, and because of the diversity of physical forces (temperature especially), mid-sized rivers are predicted to have the highest biological diversity.

In larger rivers (orders 6 and above), temperature and hydraulic changes are buffered by the large volume of water. Leaf litter inputs are minor due to large channel width, but primary production is limited by turbidity. As a result, the main energetic inputs to these rivers are from upstream processing of CPOM (fragments and feces), and collectors are the dominant macroinvertebrate functional feeding group.

A corollary to the RCC is the serial discontinuity concept (Ward and Stanford 1983), which addresses the effects of dams on rivers. According to this concept, dams cause a longitudinal discontinuity of physical and biological features. This should shift the continuum either up or down the stream order axis depending on the dam's location. For example, a dam placed on a mid-sized river should stabilize temperatures and flows downstream, reducing the biological diversity that was originally maintained by the physical diversity. A dam on a large river should reduce turbidity downstream from the

dam, resulting in more autochthonous production, and causing the river to function more like a mid-order river (Ward and Stanford 1983).

The river continuum concept was developed for North American forested stream ecosystems. Because it does not always accurately describe other types of lotic systems, the RCC has come under criticism since its inception. New Zealand workers first pointed out that in areas where there are no significant riparian inputs, such as desert, deep canyon, or prairie streams, the RCC does not adequately describe the structure and function of these systems. Winterbourn et al. (Winterbourn et al. 1981) showed that, in contrast with the RCC, stream invertebrates in New Zealand systems show little longitudinal shift in functional group dominance, and attribute this to the geomorphological and landscape-level differences in New Zealand streams compared to North American streams. The lack of retention of CPOM in the typically high gradient streams of New Zealand, along with the climatically unpredictable nature of these streams has resulted in a lack of shredders along with a macroinvertebrate fauna that is functionally flexible with asynchronous life histories (Winterbourn et al. 1981). However, in a later paper, Cummins (1988) noted that this was an exception, which reflected various degrees of alteration from the aboriginal condition. Minshall et al (1983) found that watershed climate and geology, riparian conditions, tributaries, and other location-specific factors can also cause a river to deviate from its predicted position along the river continuum based on what might be initially expected based on stream order alone. In many situations, the RCC holds, and in situations where conditions deviate from what is predicted, the RCC still serves as a useful paradigm for understanding lotic ecosystems (Cummins 1988; Minshall et al. 1985).

Perhaps the most serious questioning of the RCC has come from work on large floodplain rivers. The RCC predicts that much of the energy fueling large river production will come from upstream processing of CPOM. Some authors maintain that the river continuum concept underestimates the importance of the floodplain in providing energy to the system (Sedell et al. 1989). The RCC was tested on several large rivers: The Moisie and Salmon Rivers are ninth-order rivers with constrained channels and no floodplain. Both rivers exhibited carbon flow characteristics that closely adhered to the RCC (Sedell et al. 1989). However, the Amazon and Parana-Plata Rivers are large, tropical rivers with extensive floodplains. These rivers showed differences in carbon processing depending on whether or not areas had close associations with the floodplain. River areas with the most interactions with their floodplains were the most productive, indicating a substantial energetic input from floodplain areas (Sedell et al. 1989). These data show that the RCC holds for large rivers confined to their banks, but that river-floodplain interactions can seriously disrupt predictions of the river continuum concept.

Flood Pulse Concept

The flood pulse concept introduces a lateral dimension to the theory of lotic ecosystems. The flood pulse concept applies to large, floodplain rivers in temperate and tropical areas, and states that the most important hydrologic feature of large rivers is the predicable flooding of the river over its banks (Junk et al. 1989). Floodplains are highly productive, typically contain a wide variety of aquatic habitats, and are periodically and predictably inundated.

During a flood, aquatic organisms migrate onto the floodplain to use the available recourses. As floodwaters recede, nutrients and organic matter are funneled back into the channel, and this replenishes the resources depleted from the system since the last flood. Because the flooding is often predictable, biological communities show adaptations to using the floodplain resources (Johnson et al. 1995a;Junk et al. 1989). For example, in rivers that regularly flood, fish species, via environmental cues such as temperature or day length, anticipate the floods and spawn before or during the rise of water levels (Bayley 1995). As a result of this close tie between flooding and biological production, the flood pulse concept also predicts that hydrologic alteration, such as impoundments and channelization, along with watershed land use changes, can deleteriously affect biological communities (Bayley 1995;Johnson et al. 1995b). This is well illustrated by the channelization and impoundment of the Kissimmee River in Florida, along with the subsequent drainage and conversion of floodplain habitat for intensive agriculture (Toth 1990).

While both longitudinal (RCC) and lateral (flood pulse) attributes of lotic ecosystems are important in determining their structure and function, it is really a hierarchical combination of large scale and small-scale processes that define the structure and function of large river systems. Large-scale processes such as plate tectonics (influencing underlying geological features) and climate (affecting rainfall and flooding) tend to influence river morphology and species pools. Within these higher levels of organization, smaller-scale processes such as species interactions (competition and predation) and flow characteristics (influencing substrate particle size) operate to give

each river its characteristic population-level, community-level, and ecosystem-level structure (Johnson et al. 1995a).

ANTHROPOGENIC IMPACTS ON LARGE RIVER ECOSYSTEMS

There are few pristine large river ecosystems remaining in the world. Agricultural and urban development along river corridors have had serious impacts to large river systems related to clearing the riparian area, and the subsequent loss of retention of nutrients, sediments, and toxins, along with a reduction in the overall terrestrial inputs of CPOM and large woody debris (LWD). Hydrologic alterations, in the form of impoundments and channelization can reduce interaction between the river and its floodplain (Stanford 1996). The following is a brief discussion of the impacts humans have had on large river systems.

Watershed Land Use

The conversion of land for intensive agriculture or urban development has had both direct and indirect effects on lotic ecosystem function (Allan and Flecker 1993). Forested watersheds generally act as filters for the stream channels, buffering sediment and nutrient loads, and capturing toxins before they enter the river (Karr 1991). Riparian cover also buffers temperature changes, resulting in lower maximal values in the summer and higher minimal values in the winter (Allan and Flecker 1993). When a forested watershed is converted for agriculture, this buffering capacity is lost. Nonpoint source pollutants such as suspended sediments (Wolman 1971) and nutrients (Smith et al. 1987) generally increase, and the addition of pesticides and herbicides to the watershed often

have adverse effects on stream metabolism (Young and Huryn 1999). Urban development, specifically the addition of roads and other non-porous substrates to the watershed, increase the overland flow of water into the river, also resulting in increased sedimentation (Karr 1991) and fossil fuel runoff. Another way in which urban development has affected large river systems is through atmospheric deposition of toxic substances such as PCBs and metals (Smith et al. 1987).

Hydrologic Alteration

Hydrologic alteration of rivers has also had serious impacts on large river systems. As discussed above, the serial discontinuity hypothesis (Ward and Stanford 1983) addresses the effects of dams on the river continuum. Dams can also affect the flood pulse (Johnson et al. 1995a). By controlling the release of impounded water, water level variation below the dam can be substantially reduced. This has overall impacts on water temperature and dissolved oxygen (depending on whether the dam is "surface release" or "deep release"), as well as sediment and nutrient retention, which can affect most of the river's important ecological processes (Ligon et al. 1995).

Channel alteration, including complete channelization has been shown to have deleterious effects on river structure and function. In the late 1960's, the U.S. Army Corps of Engineers began channelization of the Kissimmee River in Florida for the purpose of flood control and floodplain development. Since channelization, there has been a 90% decrease in wading bird and waterfowl populations (Weller 1995), along with a decline in the once outstanding largemouth bass fishery (Merritt et al. 1996). The loss of littoral fringe habitat in the main channel reduced the available habitat for

macroinvertebrates and fishes, and the reduction of flow through the remnant channel resulted in a shift in the invertebrate community to one more characteristic of lentic systems (Merritt et al. 1996; Merritt et al. 1999).

ASSESSING IMPACTS ON LARGE RIVER ECOSYSTEMS

In order to address the threats to lotic ecosystem integrity discussed above, there must be an objective means of monitoring changes in river health and quantifying effects of anthropogenic effects. Using living organisms to evaluate water quality has its roots in the concept of the saprobian system (Cairns and Pratt 1993). This idea builds on the concept that certain organisms, because of their differing tolerances toward organic enrichment, could be used as indicators of ecosystem stress. This concept of indicator organisms is still used today (Hilsenhoff 1987; Hilsenhoff 1988). However, as more was learned about stream ecology and aquatic insects, problems with the saprobian indicator concept became apparent. Most aquatic insects have restricted distributions or seasonal fluctuations, which preclude their use as indicators except in specified areas where they occur and during the season in which they are most likely to be captured as part of any sampling protocol. These problems are compounded with the diversity of hydrologic and habitat characteristics of any given stream or river. Additionally, the lack of solid autecological information on most species makes their use as indicators tenuous at best, and dangerous at worst (Cairns and Pratt 1993).

Because of these problems, community measures of ecosystem integrity were introduced within the field of bioassessment. These provide information on community structure that goes beyond simple indicator species. Commonly, these types of indices

are manifested in the form of the diversity index. The Shannon Diversity Index is perhaps the most widely used of these. Generally, a diversity index incorporates some measure of total taxa richness combined with the relative representation of each taxon.

James Karr and others (Fore et al. 1996; Karr 1987; Karr 1999; Karr and Chu 1999; Reynoldson et al. 1997; Thorne and Williams 1997) developed the idea of the multimetric index, which incorporates population measures (such as indicator groups) along with community measures (such as diversity). Each metric is scored based on benthic invertebrate samples from the area to be assessed, and individual metric scores are combined to give an overall assessment of the stream, river, or lake that is being evaluated. Currently, such a scoring system is used by the Michigan Department of Environmental Quality (MDEQ) for wadeable stream biomonitoring (MDNR 1991).

Multivariate bioassessment protocols, which evaluate ecosystem health based on an expected biota are gaining wider acceptance in the field of biomonitoring (Hawkins et al. 2000;Moss et al. 1999;Wright 1995). The observed community (from the samples) is then compared to the expected community (derived from reference areas), and assessment is based on the difference. The main problem with multivariate methods is the taxonomic resolution required for such studies (Cao et al. 1996). While much of the state and federal programs in the U.S. require only family-level identification of macroinvertebrates, multivariate methods often require species-level identification. As mentioned above, multivariate assessments require the use of reference sites to set initial expectations for benthic communities. This is difficult with non-wadeable rivers because of the size of the watershed and the historical usage of these rivers for irrigation, logging,

and transportation—there simply are no large rivers in Michigan that have not at one time been impacted by human use.

Another approach to monitoring lotic systems has been proposed by Merritt et al. (Merritt et al. 1996;Merritt et al. 1999), which builds on the concepts of macroinvertebrate functional groups (Cummins and Klug 1979) and the RCC (Vannote et al. 1980). This approach uses macroinvertebrate functional group ratios to assess ecosystem function. The premise is that aquatic invertebrate communities predictably respond to changes in their physical environment, and these changes are reflected in the community-wide functional group representation. This type of assessment involves constructing macroinvertebrate functional group ratios that serve as analogues to ecosystem parameters. For example, the ratio of autochthonous to allochthonous inputs, or the ratio of photosynthetic production to respiration (P/R) can be approximated by the ratio of live vascular plant shredders + scrapers as a proportion of CPOM detritivorous shredders + total collectors (Merritt et al. 1996). Habit and voltinism groups can also be used as ecosystem analogues. For instance, to obtain a measure of habitat stability, one can use the ratio of clingers + climbers as a proportion of burrowers + sprawlers + swimmers (habit groups) or species with > 1 generation per year as a proportion of species with ≤1 generations per year (voltinism groups) (Merritt et al. 1996). The use of functional group analogues to evaluate ecosystem function and stability has been successful in the Kissimmee (Merritt et al. 1996;Merritt et al. 1999) and Caloosahatchee (Merritt et al. 2002) Rivers in Florida.

In general, biological assessment protocols commonly use a variety of assemblages to infer ecological condition. Fish (Jennings et al. 1995) and

macroinvertebrates (citation) are generally the most common assemblages used, but benthic algae communities have recently received attention in the literature and are especially useful due to the extensive autecological knowledge of algae. Because each has its advantages and disadvantages relating to temporal and spatial responses to anthropogenic stressors, many protocols use multiple assemblages in ecological assessments in addition to physical and chemical parameters important to the system of interest.

Currently, there are many state and federal biomonitoring programs that use macroinvertebrates to evaluate the integrity of streams. Most of these programs have been developed solely for smaller, wadeable streams. Because of the inherent differences between wadeable and non-wadeable river structure and function, these methods are not suitable for non-wadeable river assessment in Michigan.

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CHAPTER 2

SPATIAL AND TEMPORAL VARIATION IN THE PHYSICAL AND CHEMICAL ASPECTS OF NON-WADEABLE RIVERS IN MICHIGAN

CHAPTER 2

SPATIAL AND TEMPORAL VARIATION IN THE PHYSICAL AND CHEMICAL ASPECTS OF NON-WADEABLE RIVERS IN MICHIGAN

INTRODUCTION

Water quality and habitat quality are intimately linked with macroinvertebrate diversity, life history, and growth. Some of these parameters change predictably along the river continuum (e.g., turbidity) in a natural manner (Vannote et al. 1980), and others are influenced by seasonal factors like flooding (Junk et al. 1989). Invariably, anthropogenic influences shape the physical and chemical traits of lotic ecosystems, and these influences manifest themselves on multiple spatial and temporal scales. While much work has been done on the relationships between physical aspects of lotic environments and the associated biota, these relationships are poorly understood in non-wadeable rivers because of their large watersheds and corresponding multitude of processes that determine a non-wadeable river's physico-chemical signature. When undertaking a reach scale bioassessment project such as the one in the following chapter, it is important to discern the spatial and temporal variability of these parameters at the reach scale, especially when determining stressor-response relationships.

Water quality is ultimately reflected in the macroinvertebrate communities, and this presents several advantages compared to simply measuring these parameters directly. Macroinvertebrates are useful indicators of water quality because they are present in almost every aquatic environment. They are also indicative of environmental quality at the local scale, as compared to fish, which are much less sedentary in nature. Unlike

organisms with shorter life cycles (e.g., algae), they also allow the effects of regular or intermittent perturbation to be detected (Resh et al. 1996). However, the synergistic effects of various stressors any given macroinvertebrate population or community encounters throughout its life cycle can make it more difficult to isolate the effects of any one stressor by studying them in their natural environments. Macroinvertebrates do, however, lend themselves nicely to experimental studies. This allows information from monitoring studies such as this one to be tested and causal mechanisms to be analyzed (Resh et al. 1996). The following is a short review of some of these parameters' effects—both direct and indirect—on large river macroinvertebrates.

Temperature is one of the most important variables for river biota, affecting macroinvertebrates in many different ways. A direct effect of temperature is on metabolic rates. Like all exothermic organisms, macroinvertebrate growth rates are determined largely by temperature. Temperature also affects solubility of gases in water such as oxygen. Typically, the greatest source of heat in large rivers is direct solar radiation, since most their surfaces are directly exposed to sunlight.

Conductivity is the measure of electrical conductance of water, and an approximate indicator of dissolved ions. Variation in conductivity depends on the relative influence of underlying geological features and precipitation in providing the system with dissolved ions, with overall discharge and evaporation as secondary influences (Allan 1995). Studies have shown that stream organisms require water of some minimal ionic concentration (Willoughby and Mappin 1988), and periphyton productivity has been shown to be higher in waters with more dissolved ions (Hill and Webster 1982). In general, water of very low ionic concentrations appears to support a

reduced fauna, and the number of species commonly increases with ionic concentrations (Allan 1995).

Another physical aspect of non-wadeable rivers of importance to macroinvertebrates is pH. The acidity of water has been shown to increase macroinvertebrate drift and survival, with mayflies showing the most significant sensitivity (Courtney and Clements 1998). This factor has also been shown to influence leaf pack processing rates, with neutral streams showing the fastest rates, alkaline streams with intermediate rates, and acidic streams with the slowest rates (Griffith and Perry 1993). This effect can be direct (inhibition of macroinvertebrates themselves) or indirect due to the inhibition of microbial activity on the leaf surface (Griffith and Perry 1993; Tuchman 1993). While the range of pH in most Michigan rivers is not in itself wide enough to cause direct damage to macroinvertebrate communities (Schell and Kerekes 1989), there are other important indirect relationships between pH and possible effects on macroinvertebrates. For example, in areas with heavy metal pollution, metal dissolution in the water column has been shown to be heavily influenced by pH, which may have direct effects on macroinvertebrates or cascading effects caused by algal sensitivity to metals (Aliotta et al. 1983; Kozitskaya and Komarenko 1995; Peterson et al. 1984).

Dissolved oxygen (DO) also greatly affects aquatic life. The amount of dissolved oxygen in water bodies is influenced by abiotic aspects of the system such as temperature and barometric pressure, as well as biotic processes like respiration (decomposition) and photosynthesis. Low DO is often a result of nutrient enrichment stemming from agricultural runoff in streams, and this is commonly what is targeted in multimetric indices. In addition to nutrient enrichment, DO levels may decline to dangerous levels in

areas with high quantities of organic matter. Macroinvertebrates have differing tolerances to low DO, and the relative abundance of tolerant groups is often used as a means of evaluating organic pollution in small streams (Hilsenhoff 1987;Hilsenhoff 1988).

Fine sediments in the water column can also influence macroinvertebrate communities. Turbidity increases with riparian clearing, increases in agricultural landuse, as well as an increase in impervious substrates such as roads due to urban development and construction projects in the watershed (Wood and Armitage 1997). When this sediment settles to the bottom, it causes embeddedness of coarser substrates, which is detrimental to some fish species (Turnpenny and Williams 1980). Suspended as well as benthic fine particulate organic matter (FPOM) can also result in shifts in functional feeding groups of macroinvertebrates. For example, high suspended sediment loads may inhibit filtering collectors by clogging nets (e.g., Hydropsychidae) (Lemly 1982) or scrapers clingers (Kaller 2002) by covering hard substrates such as boulders, rocks, or large woody debris (LWD). Increased suspended sediments has also been documented to reduce macroinvertebrate density, biomass, as well as EPT taxa richness (Angradi 1999).

As mentioned above, nutrient enrichment is often the focus of environmental monitoring studies because of the effects it has on dissolved oxygen levels. In a nutrient limited system like small streams, enrichment of nitrogen or phosphorous causes algae to bloom out of control. These algae eventually die and settle to the bottom where decomposers begin to break them down. Because these decomposers are heterotrophic (e.g., bacteria), oxygen levels begin to decline rapidly as a result of increased biological

oxygen demand. In many cases, DO declines to levels that are lethal to the more sensitive groups of macroinvertebrates and can also result in fish kills. Nutrient enrichment is primarily caused by intensive agriculture in the watershed and is facilitated by the clearing of riparian vegetation that would normally buffer nutrient inputs.

All of the parameters discussed above act to influence the algal community—both suspended and benthic. Algae need light to photosynthesize, so they are influenced by suspended solids. Indeed, most large rivers are primarily limited by light because they are deeper and have much higher suspended sediment loads than small streams (Vannote et al. 1980), and this phenomenon is often seasonal in nature (Knowlton and Jones 1996; Koch et al. 2004). Aside from the effects of suspended and benthic algae have on DO levels, these communities are also important food sources for filtering collectors and scrapers.

The factors discussed above work in concert with habitat parameters such as availability of large woody debris (LWD), substrate size, riparian vegetation, and hydrologic variation to shape lotic macroinvertebrate communities, and a large-scale study of Michigan's non-wadeable river physical and chemical properties has never been done that describes these properties. The objectives of this study were to 1) describe the range of physical and chemical parameters in Michigan's non-wadeable rivers; 2) determine which parameters are common to entire catchments and which are more descriptive of reach scale properties; and 3) determine the year to year variability in these parameters from reaches visited multiple times.

METHODS AND MATERIALS

I used a YSI 6600 multiparameter data sonde to record temperature, pH, conductivity, dissolved oxygen, turbidity, and suspended chlorophyll at each study reach (Table 3.1). Readings were taken at each transect in the vicinity of macroinvertebrate sampling. The sonde was calibrated daily for dissolved oxygen, turbidity, and chlorophyll. I used barometric pressure to calibrate the oxygen probe to 100% saturation. In the field, the turbidity and chlorophyll probes were calibrated daily using distilled water. Turbidity was also calibrated weekly in the laboratory using a turbidity standard (100 NTU). I calibrated pH on a weekly basis using the 3-point method (pH=4,7,and 10). Conductivity was also calibrated weekly (conductivity=0 mS/cm and 100 mS/cm).

Another sonde was deployed at most sites for a period of 24h and set to log environmental data every 15 minutes. This sonde was used to look at daily changes in dissolved oxygen. This was done only in the 2001 sampling year, and the DO probe malfunctioned for many of the sites, and as a result, diel oxygen data are only available for 10 sites: sw_sg, sg_zil, ra_dun, mk_trk, mk_thp, kz_ver, kz_cus, gr_ion, gr_gr, and gr_cmr (see Table 3.1 for site codes).

Nutrient Samples

Water samples were taken at transects A, G, and K (Figure 3.1) at each site by placing 250 mL Nalgene bottles below the surface of the water, allowing water to flow into the bottle. Each time, I rinsed the bottles with river water 3 times prior to capping the bottle. Bottles were acid washed by soaking them in 70% H_2SO_4 for 48 hours prior to use. In the field, bottles were placed on ice in a large cooler. At the end of each day, all

bottles were returned to the lab and samples were frozen until they were sent to Michael Grant (analytical chemist, UMBS). Each sample was analyzed for nitrate, ammonia, total N, SRP, and total P.

Chlorophyll Samples

Periphyton was sampled by scraping 2 10cm^2 subsamples from large rocks (at sites with cobble) or large woody debris. Periphyton was scraped with a toothbrush and rinsed into the calibration cup of the YSI 6600 multiparameter sonde with distilled water so the total volume of the sample was 200mL. Chlorophyll was measured with the sonde in the total sample before filtering a 25 mL subsample through a Whatman GFC filter paper in the field to be used for actual chl a analysis. Samples were taken at transects A, G, and K.

I sampled phytoplankton with a standard plankton net by taking five sweeps of the net through the water column (in order to make sure algal concentrations would be high enough for chl a analysis). The sample was rinsed into the collection jar at the bottom of the net and poured into the sonde calibration cup. At this point, I recorded the sonde chlorophyll readings, and then filtered a 50 mL subsample through a Whatman GFC filter paper to be used for actual chl a analysis. Samples were taken at transects A, G, and K.

In both cases, filter papers were placed in covered plastic petri dishes and wrapped in aluminum foil in the field before they were placed on ice. At the end of each day, all chlorophyll samples were frozen for chlorophyll a extraction analysis. The actual

values (from chl a analysis) were then compared to the sonde values in order to evaluate the accuracy of the chlorophyll probe with linear regression.

I analyzed the sonde data by performing an ANOVA on all sites using individual transect data as replicates. This allowed the coarse evaluation of overall differences among sites for each parameter. To examine differences among sites on the same river I performed separate ANOVAs on sonde measurements using only values from sites within the same catchment (e.g., Grand River) and within the same sampling year (e.g., 2001 or 2002) in order to isolate catchment vs. year to year variation in values. Year to year variation in physical parameters was analyzed using ANOVA on sites that were repeated each year. See Table 3.1 for a list of sites and corresponding sampling dates. I also used ANOVA to examine differences in nutrients, phytoplankton, and periphyton among sites from 2001 only. My 2002 samples were subjected to thawing before they were sent for nutrient or chlorophyll a analysis, and were therefore inaccurate. Year-to-year differences in sonde parameters were examined by performing individual T-tests on reaches that were sampled in both years. See Tables 2.1 and 2.2 for average values for parameters measured at each site.

RESULTS

Overall Range of Physical and Chemical Parameters

All of the sonde parameters were highly variable among sites (Table 2.3), and significant differences ($p<0.05$) were detected in all cases (Table 2.4). In addition, nutrient concentrations as well as phytoplankton and periphyton all showed overall differences at the reach scale (Tables 2.5-2.6). Temperature ranged between 19C and

30C, while conductivity ranged between 0.001-0.999 mS/cm. Michigan's non-wadeable rivers also ranged in pH, although none of the sites sampled were acidic in nature. Dissolved oxygen also showed a wide range in overall values (Table 2.3). Turbidity and suspended chlorophyll, however, showed the widest range in values across sites (Table 2.3). The results of the regression analysis showed significant relationships between sonde values and measured values of chlorophyll a for both periphyton and phytoplankton (Figure 2.1). Since temperature is confounded with time of day as well as time of year, I believe that any real differences in temperature are not necessarily biologically relevant or the result of anthropogenic impacts. For this reason, differences in average temperature will not be discussed in this chapter.

Differences Among Sites from the Same Catchment

Overall, most sonde parameters varied significantly at the reach scale in 2001 (Table 2.4). In the Grand River basin, all parameters showed differences among sites except for turbidity, which was highly variable. The Kalamazoo River sites varied at the reach scale in suspended chlorophyll, DO, and turbidity, but there were no significant differences between these two sites in neither conductivity nor pH. The two sites on the Manistee River sampled in 2001 showed no differences in suspended chlorophyll, but all other parameters were significantly different. All parameters were significantly different between the two Muskegon River sites except pH. The two sites on the River Raisin showed significant differences among all physical variables (Table 2.7). Overall differences in phytoplankton and periphyton densities were detected (Table 2.6), but because of the high correlation between actual chlorophyll a from extraction methods and

the sonde values (Figure 2.1), only sonde values will be discussed for the remainder of this chapter. See Figures 2.2-2.4 for actual differences in sonde parameters among 2001 sites.

Overall differences in nutrient concentrations were detected among sites (Table 2.5). However, these differences in dissolved nutrients among sites in the same watershed were not so found to be statistically significant, and this is likely due at least in part to the smaller number of nutrient samples taken at each site (n=3) compared to the sonde measurements (n=11). Sites within the same basin almost always had nutrient concentrations that were approximately the same. The exception to this was the Grand River. Except for ammonia and SRP concentrations, nutrients levels were significantly different among the 4 sites. The only other difference in nutrient levels between sites was in the River Raisin, which showed differences in SRP between the two sites sampled in 2001 (Table 2.8). See Figures 2.5-2.7 for actual differences in nutrient concentrations among 2001 sites.

Despite the lack of significant differences in nutrient concentrations, sites from the 2001 sampling season showed differences in both phytoplankton and periphyton as measured by the chlorophyll probe on the sonde (Table 2.6; Figure 2.8).

When basin effects were considered for 2002 sites, similar results were found. While most variables were significantly different among sites on the same river, there are exceptions. Interestingly, Grand River sites showed differences in turbidity in 2002, but showed no differences in conductivity. Similarly, the sites on the Manistee River showed no differences in turbidity in 2002. The Tahquamenon River sites were most similar

from a water quality standpoint—the only significant difference between the two sites was conductivity (Table 2.9). Nutrient data were not available for the 2002 samples.

Year-to-Year Variation in Water Quality Parameters

When differences between 2001 and 2002 values of water quality parameters were tested with individual T-tests, I found that most sites showed different water quality signatures between years. All sites that were repeated in the two field seasons showed significant differences in conductivity, with the largest difference occurring at the Grand River @ Grand Rapids (Table 2.9). All sites but the Grand River @ Johnson Park showed significant differences in pH. Dissolved oxygen levels were also different at all sites but the Grand River @ Johnson Park and the Manistee River @ High Bridge. Turbidity and suspended chlorophyll were the most similar water quality parameters between the two years. Only two sites on the Grand River (Ionia and Comstock Riverside) showed differences in turbidity between years, and one site (Grand River @ Comstock Riverside) showed significant difference in suspended chlorophyll (Table 2.9). Interestingly, all differences in conductivity and suspended chlorophyll were the results of decreases in values, but all other parameters showed some sites that increased and some sites that decreased in values from 2001 to 2002 (Table 2.9). See Figures 2.12-2.17 for actual measurements of year-to-year variation in physical and chemical values.

DISCUSSION

In general, the physical/chemical signature of non-wadeable river reaches is unique even among sites from the same catchment, suggesting that despite upstream and

catchment-wide influences, reaches maintain a unique physical environment. Most parameters were highly variable even among sites from the same catchment, and this is likely due to the many factors that influence water quality parameters in a large river ecosystem. These factors include underlying geological features, magnitude and timing of precipitation (influencing discharge), and the composition of this precipitation (Allan 1995; Castillo et al. 2000). In addition to the natural fluctuations in the physical and chemical properties of rivers, these parameters are also heavily influenced by anthropogenic influences like landuse, habitat modification, impoundment, and industrial or municipal effluents.

Conductivity appears to be highly related to geographical (geological) differences among sites, with the lowest values in conductivity occurring at the northern sites (AuSable, Muskegon, Manistee, Tahquamenon, and Menominee Rivers) in both years (Figure 2.2a and 2.9a). All sites were slightly alkaline in both years (Figures 2.2b and 2.9b), and the upper peninsula sites were close to neutral (Figure 2.9), suggesting geological differences may confer a greater buffering capacity in these sites.

Dissolved oxygen is relatively high in all sites despite some sites' heavily agricultural watersheds. This suggests the non-wadeable rivers I sampled were not nutrient limited—it is more likely they were light-limited (Knowlton and Jones 1996; Koch et al. 2004). While those sites with intensive agriculture and their associated nutrient enrichment do have higher ranges in DO (e.g., gr_ion), the minimum DO values at even these sites are not dangerously low (Figure 2.18). The sites with the lowest range in diel DO are those that are most shaded (e.g., ra_dun, kz_cus) (Figure 2.18). Notably,

the River Raisin @ Dundee (ra_dun) is also one of the most turbid sites visited in 2001 (Figure 2.3).

Both turbidity and suspended chlorophyll varied most among sites in both years (Figures 2.3b, 2.4, 2.10b and 2.11). Both parameters are highly dependant on discharge, and so the variability within each site is expected. Variability in turbidity among sites was most likely due to landuse differences (along with underlying geology) (Young and Huryn 1999). Variability in suspended chlorophyll among sites could be due to many factors including conductivity, turbidity, riparian shading, and nutrient inputs (Allan 1995).

Nutrient concentrations also varied significantly among sites (Table 2.5), and this was again likely due to differences in landuse—both catchment wide and riparian. Interestingly, some of the sites with the highest nutrient concentrations had relatively low suspended chlorophyll (phytoplankton) as well as periphyton (as measured by the chlorophyll probe). Many of these same sites also had the highest turbidity. For example, in 2001, the River Raisin @ Dundee (ra_dun01) had the highest nitrate levels (Figure 2.5b) and highest SRP levels of all sites (Figure 2.7b). Yet this site had lower suspended chlorophyll (Figure 2.4) and phytoplankton (Figure 2.8a) levels than most sites. Presumably, this is due to the fact that this site also had the highest turbidity of all sites visited in 2001 (Figure 2.3b). The site with the lowest turbidity in 2001 (ma_hbr01) (Figure 2.3b) also had the lowest suspended chlorophyll (Figure 2.4) as well as low values of all nutrients measured (Figure 2.5-2.7). This site also had low phytoplankton, but relatively high periphyton concentrations (with low standard error) (Figure 2.8). This suggests that nutrient concentrations are adequate for maintaining consistently high

periphyton levels in the absence of high suspended solids, which act to shade benthic algae.

Differences Among Sites from the Same Catchment

When assessing non-wadeable river environmental health at the reach scale, it is important that biological communities do not simply represent catchment-specific signatures. It has been shown that habitat quality varies at the reach scale (Wilhelm 2005), and these data suggest that water quality is also unique at the reach scale. Almost all physical parameters from sonde data were significantly different among sites from the Grand River and between sites from the Kalamazoo, Manistee, Muskegon, and Raisin rivers in 2001 (Table 2.7). Those parameters that were not significantly different among sites from the same basin were those that were the most variable. For example, turbidity levels were the same for all Grand River sites (Table 2.7), and this is likely due to the fact that turbidity levels within each site on the Grand River were highly variable compared to other parameters (Figure 2.3b). The Manistee River sites showed a similar pattern in suspended chlorophyll (Table 2.7; Figure 2.4). Some of these patterns may be related to proximity of sites to each other. For example, suspended chlorophyll levels in the Grand River increased from upstream to downstream (the order of these sites along the continuum, from upstream to downstream is: gr_ion, gr_cmr, gr_gr, gr_jon) (Figure 2.4). A similar pattern in chlorophyll levels is seen in the River Raisin from the upstream site (ra_dun) to the downstream site (ra_mon) (Figure 2.4). This is a common pattern related to canopy cover and discharge predicted by stream ecosystem theory (Minshall et al. 1985;Vannote et al. 1980). Similar patterns were observed in 2002, with the exception of

the two sites on the Tahquamenon River. The only significant difference in sonde parameters on this river was in conductivity (Table 2.9). The upstream site (tq_nwb02) had higher conductivity than the downstream site (tq_pds02) (Figure 2.9a).

This reach-specific distinctiveness in sonde parameters was not seen in nutrient concentrations. Almost none of the sites tested for differences in nutrient concentrations showed significant differences when compared with sites from the same catchment (Table 2.8). As mentioned above, this is probably at least partially due to the low number of nutrient samples taken at each reach (n=3). The exception to this is the Grand River, where total N, nitrate, and total P all showed differences among the 4 sites sampled (Table 2.8). One possible reason for this is the proximity of the Grand River sites to minor impoundments. Directly downstream from the Comstock Riverside site (gr_cmr), there are two small dams. This could explain why nutrient levels drop so low in this site as well as the site directly downstream from the dams (gr_gr) (Figures 2.5 and 2.7) (Castillo et al. 2000). This could also help explain the abundance of periphyton directly downstream from the dam (gr_gr). This site had the lowest turbidity of all the Grand River sites, and this is likely because the suspended solids settled out directly upstream from the dam, increasing water clarity (Ward and Stanford 1983). While most of the non-wadeable rivers in Michigan are impounded in some way, none of the other sites I visited were in such close proximity (both upstream and downstream) from a dam.

Year-to-Year Variation in Water Quality Parameters

Most physical parameters varied significantly from 2001 to 2002. In the 6 sites that were sampled in both years, conductivity was significantly different in all sites

(Table 2.10), and this was always due to in increase in this parameter (Figures 2.12-2.17). An explanation for this is the increased discharge during 2002 due to increased precipitation. The 2001 sampling season was one of the driest summers in recent years, which would have increased the concentration of dissolved ions in river waters throughout the state of Michigan. In 2002, however, there was much more rain, which diluted these ions and lowered the conductivity (personal observation). A similar pattern was found in pH from year to year. This is not surprising since pH and conductivity are highly correlated (Figures 2.12-2.17) (Allan 1995).

Patterns in DO and turbidity were not so clear. While most sites showed significant differences in DO from year to year, these values increased at some sites and decreased at others (Table 2.10). For example, mean DO levels decreased by 2.875 mg/L at the Grand River @ Comstock Riverside (gd_cmr) in 2002 as compared to 2001 (Figure 2.15). This could be due to an overall decrease in suspended chlorophyll at this site (Figure 2.15). However, at the Grand River @ Grand Rapids (gr_gr), the opposite trend in DO was observed (Table 2.9; Figure 2.12). There were also increases and decreases in turbidity levels, though none of the increases were significantly different from year to year (Table 2.9). As mentioned above, turbidity was highly variable even within each study reach.

The only site in which suspended chlorophyll levels were significantly different from year to year was the Grand River @ Comstock Riverside (gr_cmr) (Table 2.9, Figure 2.15). The chlorophyll probe malfunctioned for the Manistee and AuSable sites in the year 2002, so no data are available on year-to-year variation. See Figures 2.12-2.17 for graphical representation of year-to-year variation in sonde parameters.

Implications for the Bioassessment of Non-wadeable Rivers in Michigan

Constructing an assessment protocol of any type for non-wadeable rivers is challenging because of the large watersheds and the associated anthropogenic impacts at differing spatial scales that shape the physical and chemical environments in each reach. It is important, when undertaking such a project as developing a biomonitoring protocol for these systems, that the biological community of interest be unique at the scale at which assessment is to take place. Because biological communities in rivers are shaped by water quality and habitat quality, it is important that these factors differ at the reach scale and equally important to understand the source of this variability. This study showed that water quality parameters such as conductivity, pH, DO, turbidity, chlorophyll, and nutrients are highly variable at the reach scale, and this presumably is what causes macroinvertebrate communities to be unique at the same scale (see Chapter 3).

However, the robustness of any bioassessment protocol is of equal importance. The data reported in this study suggest that year-to-year variation in a river's physical and chemical environment is an important consideration. Indeed, this will likely be the reason for the differences in IBI scores between the two main sampling seasons since habitat at any given site changes much more slowly.

To be of most use, a dataset like this, combined with macroinvertebrate data from the same sites, could be used to formulate hypotheses that lend themselves to experimental work. This would allow actual study of mechanistic relationships between water quality (and the human behaviors that modify it) and macroinvertebrate population and community responses.

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TABLES

Table 2.1. Mean values for physical/chemical parameters by site. Parameters were recorded with a YSI 6600 multiparameter data sonde. TOT N=Total Nitrogen (ppm); TOT P=Total Phosporous (ppb); TEMP=Temperature (C); COND=Conductivity (mS/cm); PH=pH; DO=Dissolved Oxygen (mg/L); TURB=Turbidity (NTU); CHL=Suspended Chlorophyll (μg/L). See Table 3.1 for site codes.

Site Code	TOT N	TOT P	TEMP	COND	PH	DO	TURB	CHL
as_whp01	0.28	7.20	25.49	0.30	8.58	9.62	0.04	1.17
gd_cmr01	0.84	30.27	24.95	0.65	8.41	11.30	20.02	46.76
gd_gr01	0.77	29.00	25.98	0.72	8.47	10.59	27.78	40.76
gd_ion01	2.63	80.33	24.67	0.68	8.34	8.98	21.34	33.00
gd_jon01	1.90	53.00	24.94	0.68	8.50	11.51	34.62	46.88
kz_cus01	1.08	38.17	23.17	0.60	7.99	7.22	13.69	7.02
ma_hbr01	0.27	6.60	22.54	0.34	8.26	8.87	0.55	0.73
ma_rbw01	0.27	3.53	23.07	0.35	8.20	9.37	12.63	2.42
mk_thp01	0.52	19.13	22.71	0.33	8.23	9.44	0.88	5.79
mk_trk01	0.59	27.50	24.77	0.36	8.16	8.81	3.05	3.54
ra_dun01	1.52	52.35	22.40	0.64	7.97	6.36	75.41	8.46
ra_mon01	2.34	59.57	23.52	0.53	8.44	9.00	31.50	11.95
sg_zil01	1.40	32.40	27.79	0.87	8.18	5.83	38.07	15.00
sh__sg01	1.19	33.97	25.91	0.68	8.33	7.99	80.06	17.74
tb_sag01	0.74	36.10	29.52	0.88	8.80	11.51	46.18	23.48
as_mth02	0.21	9.00	27.57	0.32	8.23	8.50	N/A	N/A
as_whp02	0.13	4.50	26.01	0.30	8.07	7.46	N/A	N/A
gd_cmr02	1.48	19.07	22.40	0.57	8.17	8.42	11.12	21.46
gd_gh02	1.26	27.70	23.56	0.61	8.51	15.17	24.96	62.90
gd_gld02	1.93	37.37	N/A	N/A	N/A	N/A	N/A	N/A
gd_gr02	1.47	20.80	21.12	0.59	8.39	12.12	17.18	35.66
gd_ion02	1.42	16.53	24.37	0.56	8.15	7.74	53.05	30.87
gd_jon02	1.23	17.67	22.10	0.62	8.50	11.88	131.51	46.44
ma_cts02	0.27	4.33	22.78	0.30	8.08	9.81	N/A	N/A
ma_hbr02	0.17	5.97	24.01	0.34	8.05	9.24	N/A	N/A
ma_mns02	0.23	6.27	22.47	0.33	7.90	7.02	N/A	N/A
me_kss02	0.28	16.53	25.45	0.23	7.39	6.15	39.59	11.65
me_stb02	0.35	15.80	23.34	0.23	7.46	7.51	5.75	7.27
mk_br02	N/A	N/A	25.45	0.35	7.96	7.41	10.76	9.07
mk_nwg02	0.43	3.83	19.88	0.29	7.70	8.49	2.96	4.36
sj_mvl02	0.73	6.77	28.90	0.57	7.72	7.06	4.06	9.45
sj_rvw02	1.08	16.63	27.00	0.60	8.21	9.61	10.56	110.14
tq_nwb02	0.46	11.20	23.26	0.16	7.12	6.84	30.83	11.28
tq_pds02	0.51	6.70	22.32	0.15	7.13	6.65	9.61	11.51

Table 2.2. Mean values for landuse/habitat values by site. All values from Wilhelm (2002) except LWD. Landuse values are percentages of urban (Ur), agricultural (Ag), and natural (Nat) and are either for the entire watershed (WS) or in a 100m riparian buffer zone at each site (RIP). LWD=the number of transects with large woody debris to sample. See Table 3.1 for site codes.

Site Code	Ur WS	Ag WS	Nat WS	Ur RIP	Ag RIP	Nat RIP	LWD
as_whp01	3.67	2.34	86.29	0.00	0.00	100.00	8
gd_cmr01	7.70	59.50	23.85	93.62	0.00	6.38	4
gd_gr01	8.20	59.05	23.67	66.67	0.00	23.81	2
gd_ion01	8.66	63.56	20.08	0.00	20.00	80.00	9
gd_jon01	8.37	58.84	23.67	5.13	0.00	89.74	10
kz_cus01	8.78	55.29	28.27	0.00	0.00	95.65	10
ma_hbr01	1.81	13.31	75.83	0.00	0.00	93.18	8
ma_rbw01	1.87	13.35	74.58	6.98	0.00	93.02	9
mk_thp01	3.22	23.31	62.97	0.00	0.00	100.00	8
mk_trk01	3.32	24.95	61.52	13.16	71.05	13.16	8
ra_dun01	6.55	72.38	15.14	51.11	17.78	31.11	10
ra_mon01	6.36	74.67	13.25	54.17	0.00	31.25	1
sg_zil01	8.06	48.68	31.43	38.78	0.00	14.29	3
sh__sg01	9.56	57.13	21.49	0.00	0.00	100.00	0
tb_sag01	4.93	37.19	45.97	27.66	4.26	42.55	5
as_mth02	3.74	3.89	84.65	63.64	0.00	27.27	7
as_whp02	3.67	2.34	86.29	0.00	0.00	100.00	7
gd_cmr02	7.70	59.50	23.85	93.62	0.00	6.38	5
gd_gh02	8.62	59.09	23.31	2.22	0.00	97.78	8
gd_gld02	14.46	52.19	24.93	60.87	0.00	32.61	8
gd_gr02	8.20	59.05	23.67	66.67	0.00	23.81	3
gd_ion02	8.66	63.56	20.08	0.00	20.00	80.00	10
gd_jon02	8.37	58.84	23.67	5.13	0.00	89.74	10
ma_cts02	1.81	13.22	76.92	0.00	0.00	100.00	7
ma_hbr02	1.81	13.31	75.83	0.00	0.00	93.18	10
ma_mns02	2.05	12.09	76.06	86.36	0.00	0.00	6
me_kss02	1.94	5.74	88.35	31.82	0.00	68.18	8
me_stb02	1.90	3.16	91.29	0.00	0.00	100.00	7
mk_br02	3.37	20.56	66.07	10.64	2.13	57.45	10
mk_nwg02	3.28	23.33	63.01	8.11	16.22	70.27	10
sj_mvl02	4.25	68.24	22.99	31.48	1.85	57.41	10
sj_rvw02	5.25	66.09	23.41	61.11	2.78	27.78	7
tq_nwb02	1.28	1.95	88.95	0.00	0.00	100.00	0
tq_pds02	0.66	0.79	94.52	2.17	0.00	97.83	10

Table 2.3. Basic statistics of sonde parameters from all sites. TEMP: Temperature (C); COND: Conductivity (mS/cm); DO: Dissolved oxygen (mg/L); TURB: Turbidity (NTU); CHL: Suspended chlorophyll (ug/L)

	TEMP	COND	PH	DO	TURB	CHL
N of cases	374	374	374	374	372	319
Minimum	19.120	0.001	7.000	5.350	-1.100	-50.000
Maximum	30.400	0.999	8.880	16.230	317.300	387.500
Range	11.280	0.998	1.880	10.880	318.4	437.500
Mean	24.301	0.481	8.107	8.879	20.471	22.442
Standard Dev	2.226	0.201	0.393	2.065	34.041	36.398

Table 2.4. ANOVA results for physical parameters at all sites.

Dependant Variable	df	F-ratio	P-value
Temperature	33	149.017	<0.001
Conductivity	33	209.744	<0.001
pH	33	295.815	<0.001
Dissolved Oxygen	33	127.708	<0.001
Turbidity	33	5.401	<0.001
Chlorophyll	28	7.172	<0.001

Table 2.5. ANOVA results for nutrient concentrations for all 2001 sites.

Dependant Variable	df	F-ratio	P-value
Total N	15	7.748	<0.001
Nitrate	15	8.792	<0.001
Ammonia	15	2.968	0.005
Total P	15	7.527	<0.001
SRP	15	9.153	<0.001

Table 2.6. ANOVA results for chlorophyll concentrations for all 2001 sites.

Dependant Variable	df	F-ratio	P-value
Phytoplankton CHL	15	40.059	<0.001
Periphyton CHL	15	2.741	0.009

Table 2.7. Basin effects for sonde parameters. 2001 sites only. This table shows the results of ANOVA tests for differences among sites from the same river catchment.

River	Parameter	df	F-ratio	P-value
Grand	Chlorophyll	3	10.273	<0.001
Grand	Conductivity	3	30.381	<0.001
Grand	DO	3	19.185	<0.001
Grand	pH	3	13.330	<0.001
Grand	Turbidity	3	1.619	0.200
Kalamazoo	Chlorophyll	1	42.680	<0.001
Kalamazoo	Conductivity	1	0.070	0.794
Kalamazoo	DO	1	19.876	<0.001
Kalamazoo	pH	1	2.583	0.124
Kalamazoo	Turbidity	1	16.620	0.001
Manistee	Chlorophyll	1	1.525	0.231
Manistee	Conductivity	1	34.946	<0.001
Manistee	DO	1	5.531	0.029
Manistee	pH	1	12.744	0.002
Manistee	Turbidity	1	7.101	0.015
Muskegon	Chlorophyll	1	5.453	0.030
Muskegon	Conductivity	1	133.008	<0.001
Muskegon	DO	1	18.864	<0.001
Muskegon	pH	1	3.161	0.091
Muskegon	Turbidity	1	9.553	0.006
Raisin	Chlorophyll	1	4.635	0.044
Raisin	Conductivity	1	4.893	0.039
Raisin	DO	1	266.914	<0.001
Raisin	pH	1	363.795	<0.001
Raisin	Turbidity	1	17.455	<0.001

Table 2.8. Basin effects for nutrient concentrations. 2001 sites only. This table shows the results of ANOVA tests for differences among sites from the same river catchment.

River	Parameter	df	F-ratio	P-value
Grand	Total N	3	51.452	<0.001
Grand	Nitrate	3	35.864	<0.001
Grand	Ammonia	3	0.940	0.465
Grand	Total P	3	19.883	<0.001
Grand	SRP	3	3.485	0.090
Kalamazoo	Total N	1	0.384	0.569
Kalamazoo	Nitrate	1	0.343	0.590
Kalamazoo	Ammonia	1	0.169	0.702
Kalamazoo	Total P	1	0.186	0.688
Kalamazoo	SRP	1	1.569	0.279
Manistee	Total N	1	0.001	0.980
Manistee	Nitrate	1	0.249	0.644
Manistee	Ammonia	1	8.508	0.043
Manistee	Total P	1	3.398	0.139
Manistee	SRP	1	1.690	0.263
Muskegon	Total N	1	0.286	0.621
Muskegon	Nitrate	1	0.049	0.836
Muskegon	Ammonia	1	4.211	0.109
Muskegon	Total P	1	2.097	0.221
Muskegon	SRP	1	0.664	0.461
Raisin	Total N	1	0.550	0.512
Raisin	Nitrate	1	0.329	0.607
Raisin	Ammonia	1	0.353	0.594
Raisin	Total P	1	0.104	0.769
Raisin	SRP	1	312.006	<0.001

Table 2.9. Basin effects for sonde parameters. 2002 sites only. This table shows the results of ANOVA tests for differences among sites from the same river catchment.

River	Parameter	df	F-ratio	P-value
AuSable	Chlorophyll	1	n/a	n/a
AuSable	Conductivity	1	19.212	<0.001
AuSable	DO	1	19.935	<0.001
AuSable	pH	1	16.502	0.001
AuSable	Turbidity	1	0.889	0.357
Grand	Chlorophyll	4	63.881	<0.001
Grand	Conductivity	4	0.976	0.429
Grand	DO	4	223.180	<0.001
Grand	pH	4	181.809	<0.001
Grand	Turbidity	4	2.727	0.040
Manistee	Chlorophyll	2	n/a	n/a
Manistee	Conductivity	2	391.937	<0.001
Manistee	DO	2	168.892	<0.001
Manistee	pH	2	41.673	<0.001
Manistee	Turbidity	2	0.749	0.482
Menominee	Chlorophyll	1	6.518	0.019
Menominee	Conductivity	1	0.442	0.514
Menominee	DO	1	41.183	<0.001
Menominee	pH	1	1.358	0.251
Menominee	Turbidity	1	1.571	0.224
Muskegon	Chlorophyll	1	18.586	<0.001
Muskegon	Conductivity	1	291.267	<0.001
Muskegon	DO	1	95.095	<0.001
Muskegon	pH	1	43.773	<0.001
Muskegon	Turbidity	1	63.320	<0.001
St. Joseph	Chlorophyll	1	4.598	0.044
St. Joseph	Conductivity	1	401.117	<0.001
St. Joseph	DO	1	435.887	<0.001
St. Joseph	pH	1	316.283	<0.001
St. Joseph	Turbidity	1	45.765	<0.001
Tahquamenon	Chlorophyll	1	0.014	0.907
Tahquamenon	Conductivity	1	202.661	<0.001
Tahquamenon	DO	1	0.987	0.332
Tahquamenon	pH	1	0.342	0.565
Tahquamenon	Turbidity	1	3.735	0.068

Table 2.10. Mean differences (D) and associated p-values from individual T-tests on physical parameters between 2001 and 2002 field seasons. A negative difference indicates values decreased. A positive difference indicates values increased. N/A: Data not available. See Table 3.1 for site codes.

Site	Conductivity mS/cm		pH		DO mg/L		Turbidity NTU		Chlorophyll ug/L	
	D	p-value	D	p-value	D	p-value	D	p-value	D	p-value
gd_jon	-0.059	<0.001	+0.003	0.927	+0.363	0.280	-22.848	<0.001	-0.446	0.811
gd_cmr	-0.096	<0.001	-0.239	<0.001	-2.875	<0.001	-8.900	<0.001	-25.300	<0.001
gd_gr	-0.134	<0.001	-0.081	0.002	+1.527	0.003	-10.602	0.317	-5.104	0.192
gd_ion	-0.115	0.046	-0.195	<0.001	-1.218	0.004	+28.506	0.168	-2.583	0.409
as_whp	-0.006	0.001	-0.514	<0.001	-2.154	<0.001	+5.573	0.115	N/A	N/A
ma_hbr	-0.004	0.002	-0.217	<0.001	+0.370	0.074	+0.745	0.641	N/A	N/A

FIGURES

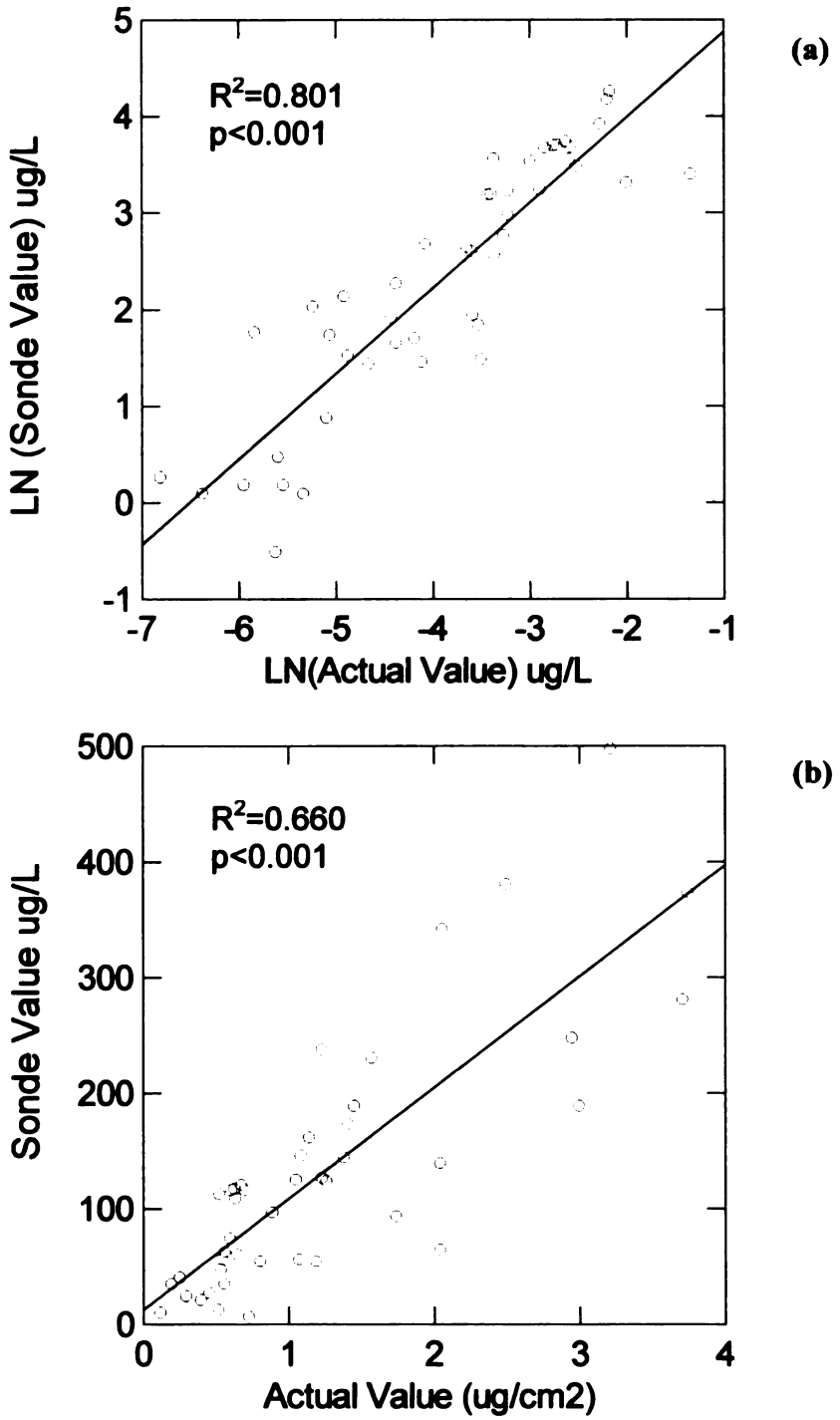

Figure 2.1. Actual values from chl a extraction vs. sonde values for (a) phytoplankton and (b) periphyton samples.

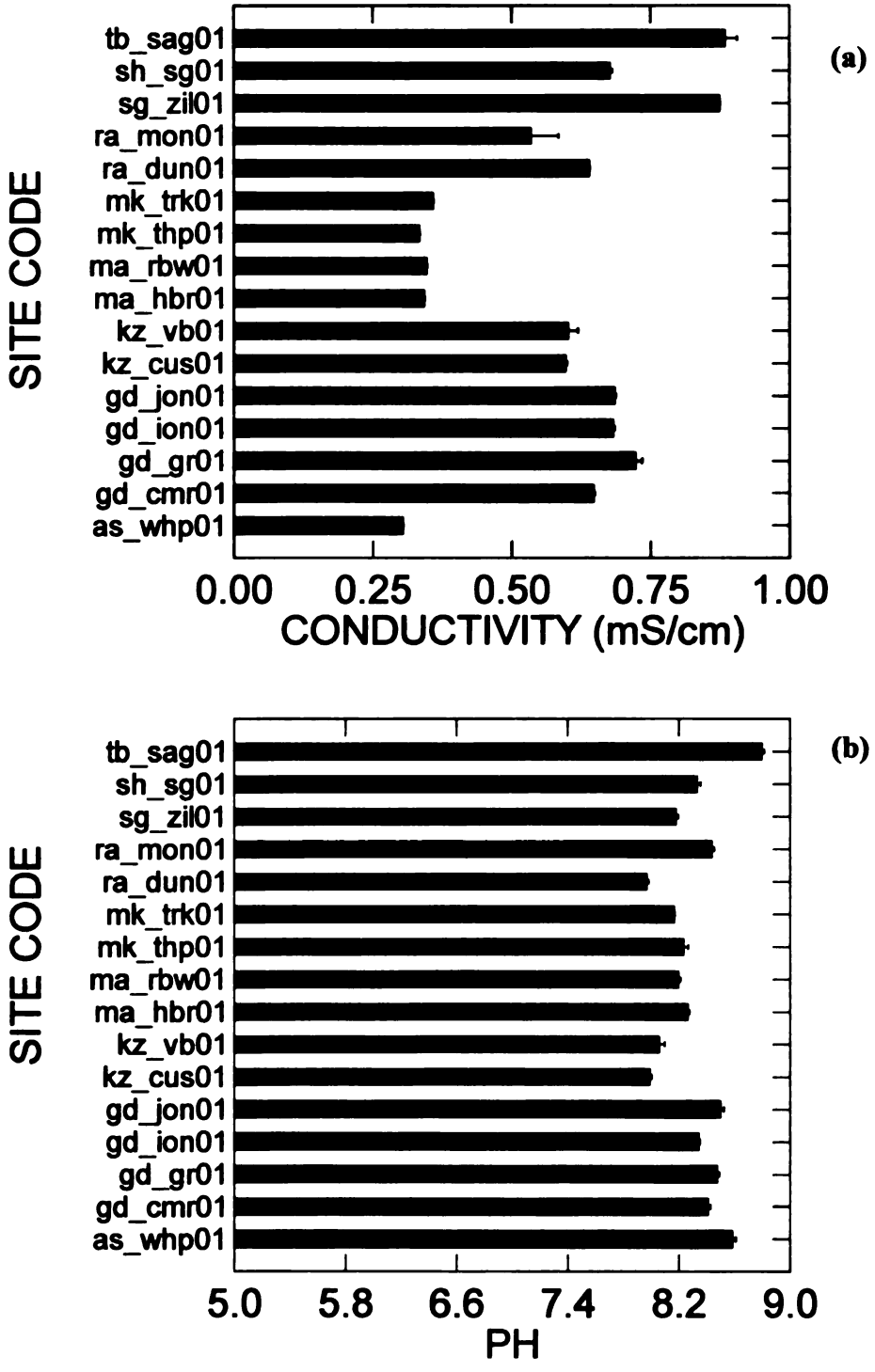

Figure 2.2. Mean (a) conductivity and (b) pH for all 2001 sites. Error bars indicate standard error. See Table 3.1 for site codes.

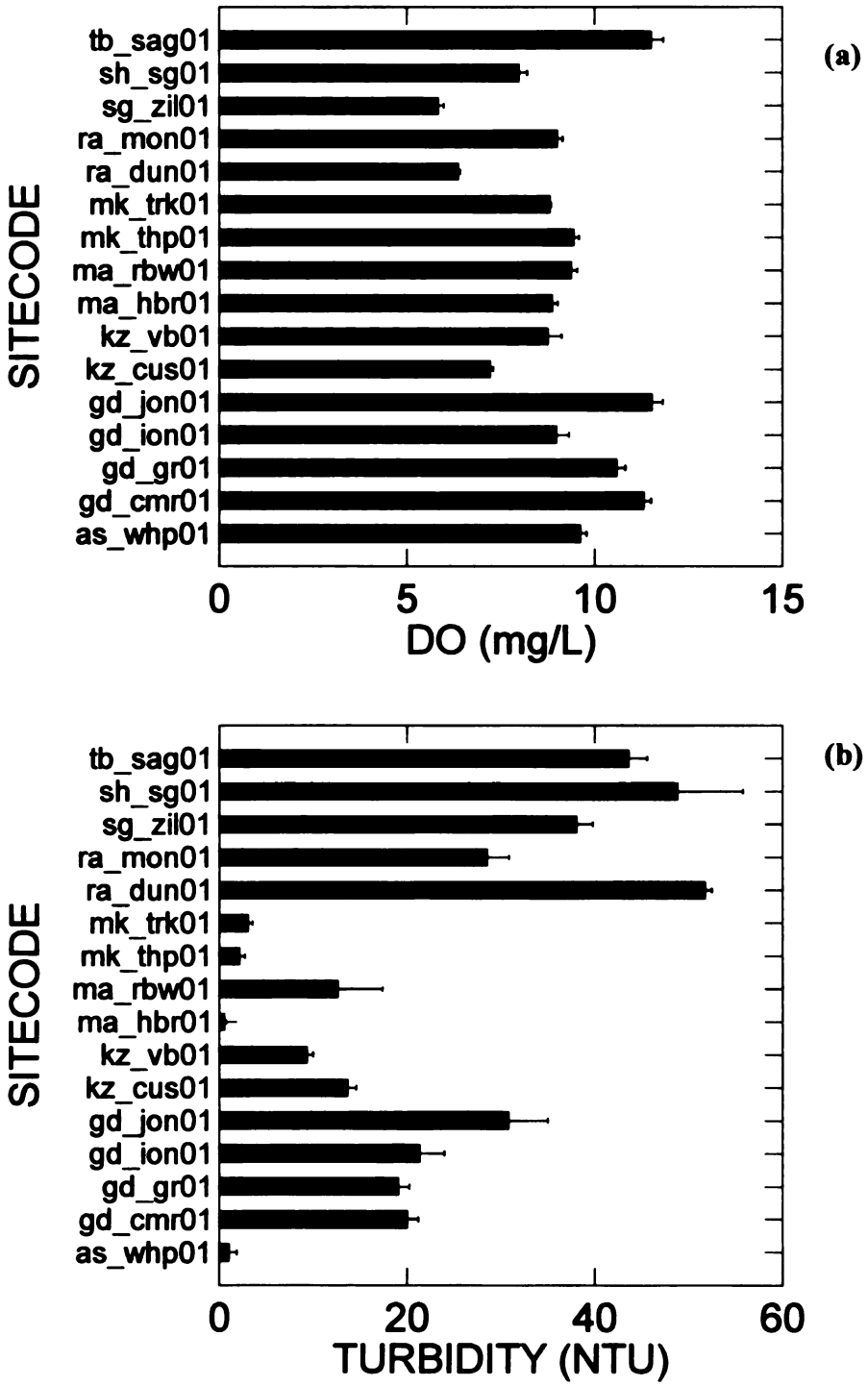

Figure 2.3. Mean (a) dissolved oxygen (DO) and (b) turbidity for all 2001 sites. Error bars indicate standard error. See Table 3.1 for site codes.

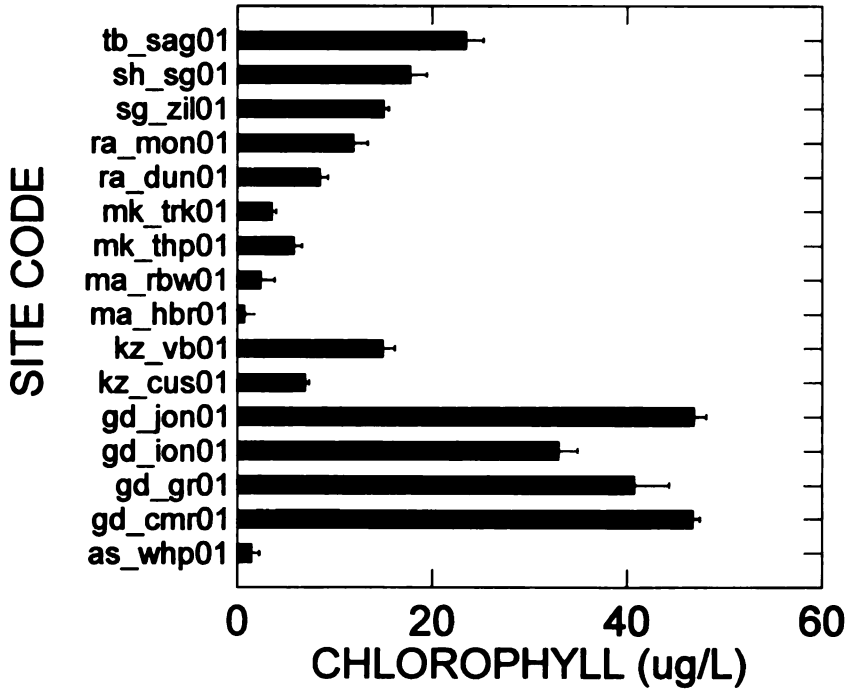

Figure 2.4. Mean chlorophyll values for all 2001 sites. Error bars indicate standard error. See Table 3.1 for site codes.

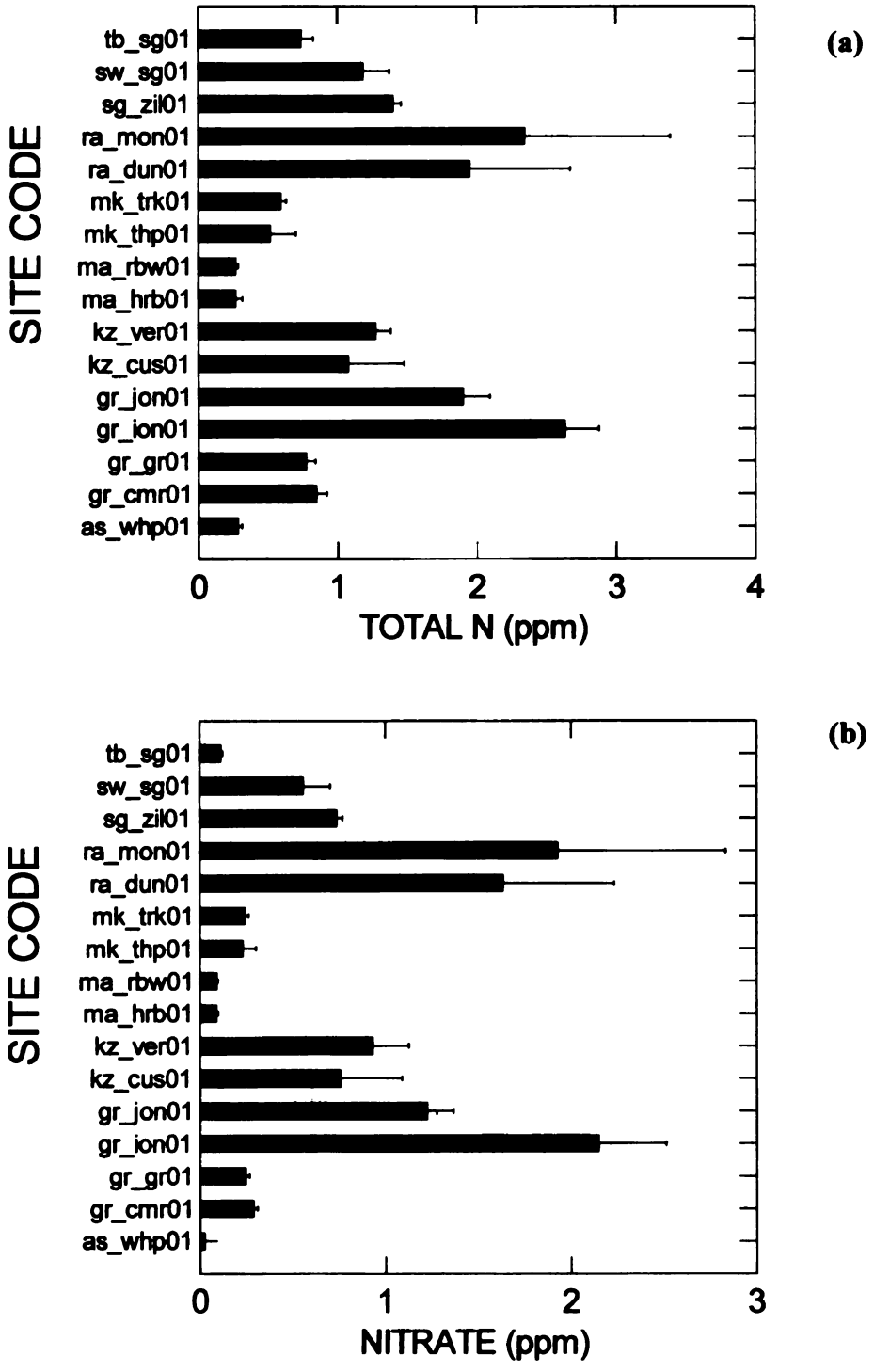

Figure 2.5. Mean (a) total N and (b) nitrate for all 2001 sites. Error bars indicate standard error. See Table 3.1 for site codes.

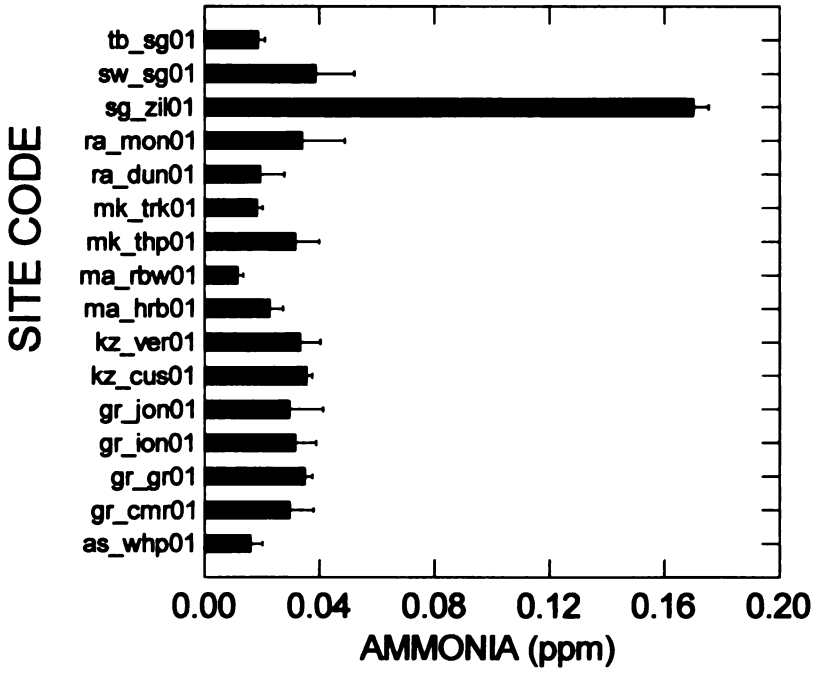

Figure 2.6. Mean ammonia for all 2001 sites. Error bars indicate standard error. See Table 3.1 for site codes.

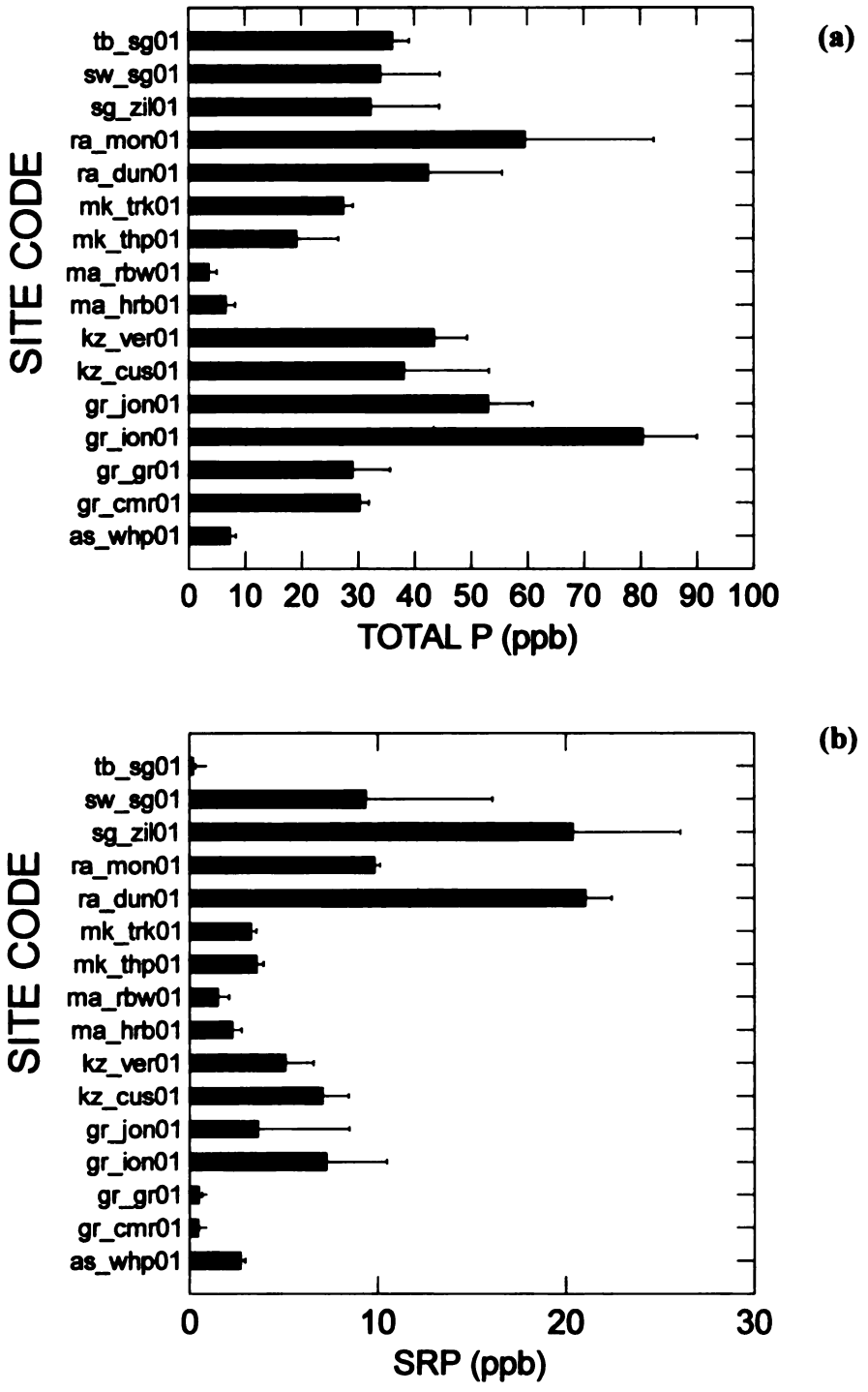

Figure 2.7. Mean (a) total P and (b) soluble reactive P (SRP) for all 2001 sites. Error bars indicate standard error. See Table 3.1 for site codes.

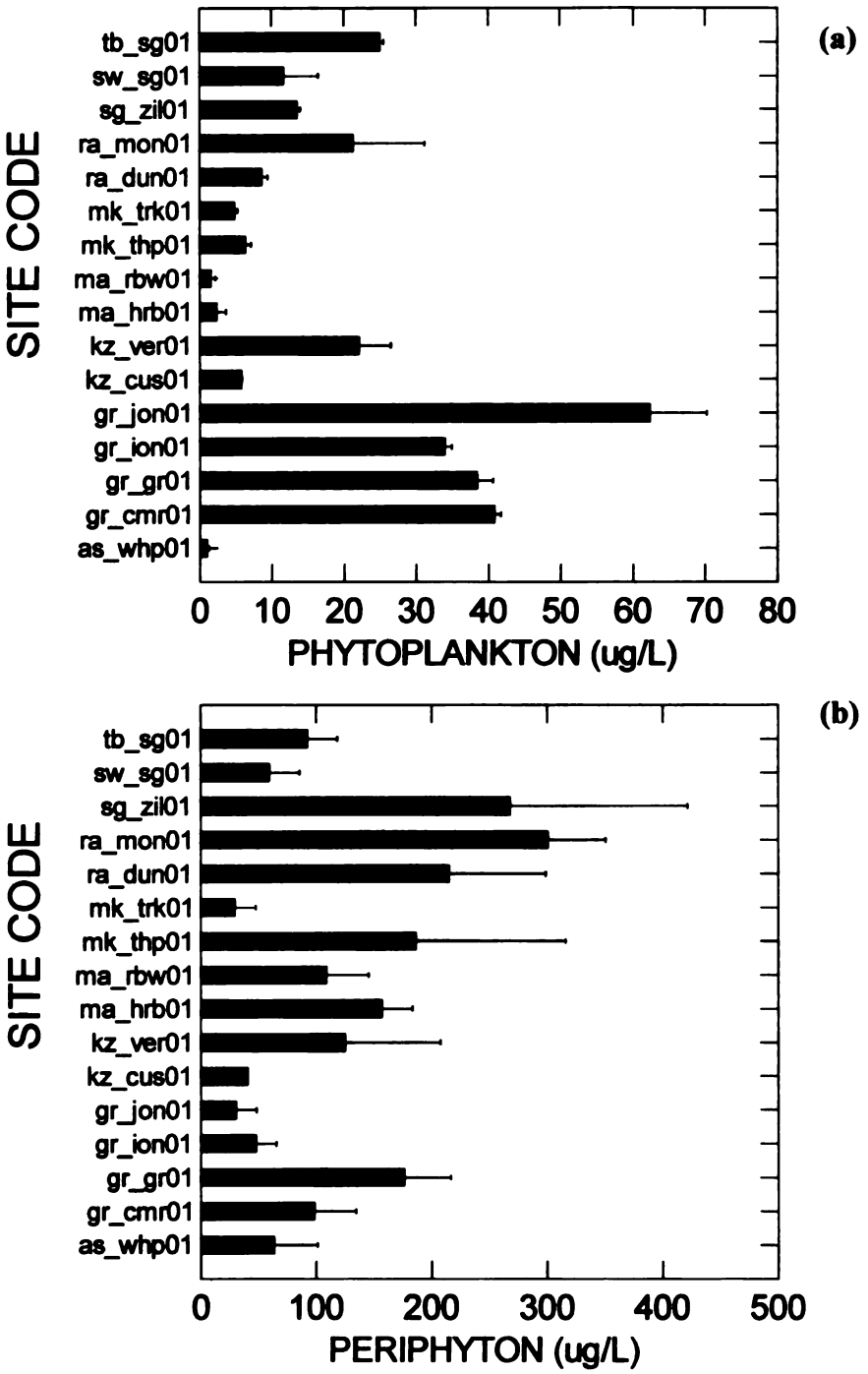

Figure 2.8. Mean (a) phytoplankton and (b) periphyton for all 2001 sites. Data are from sonde measurements. Error bars indicate standard error. See Table 3.1 for site codes.

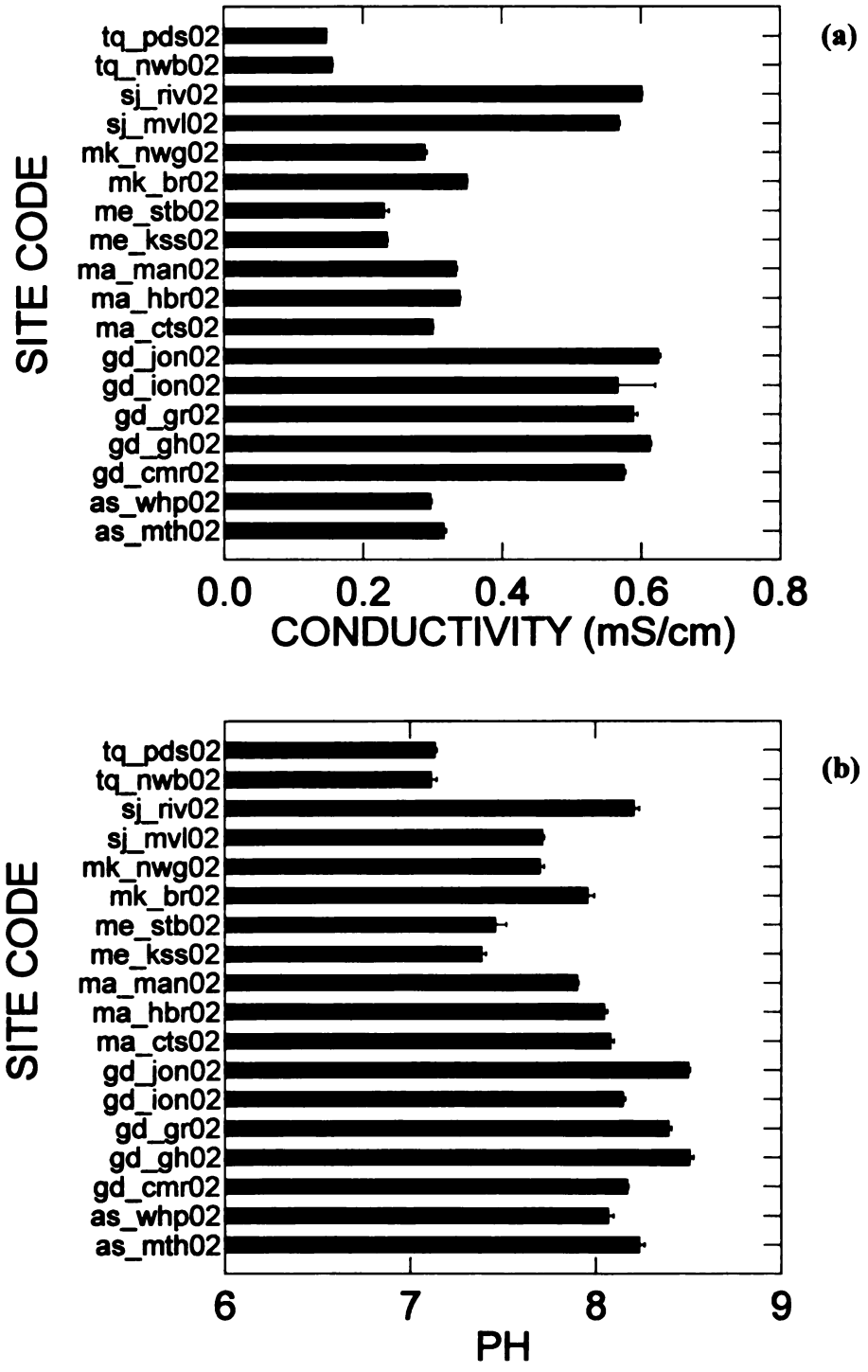

Figure 2.9. Mean (a) conductivity and (b) pH for all 2002 sites. Error bars indicate standard error. See Table 3.1 for site codes.

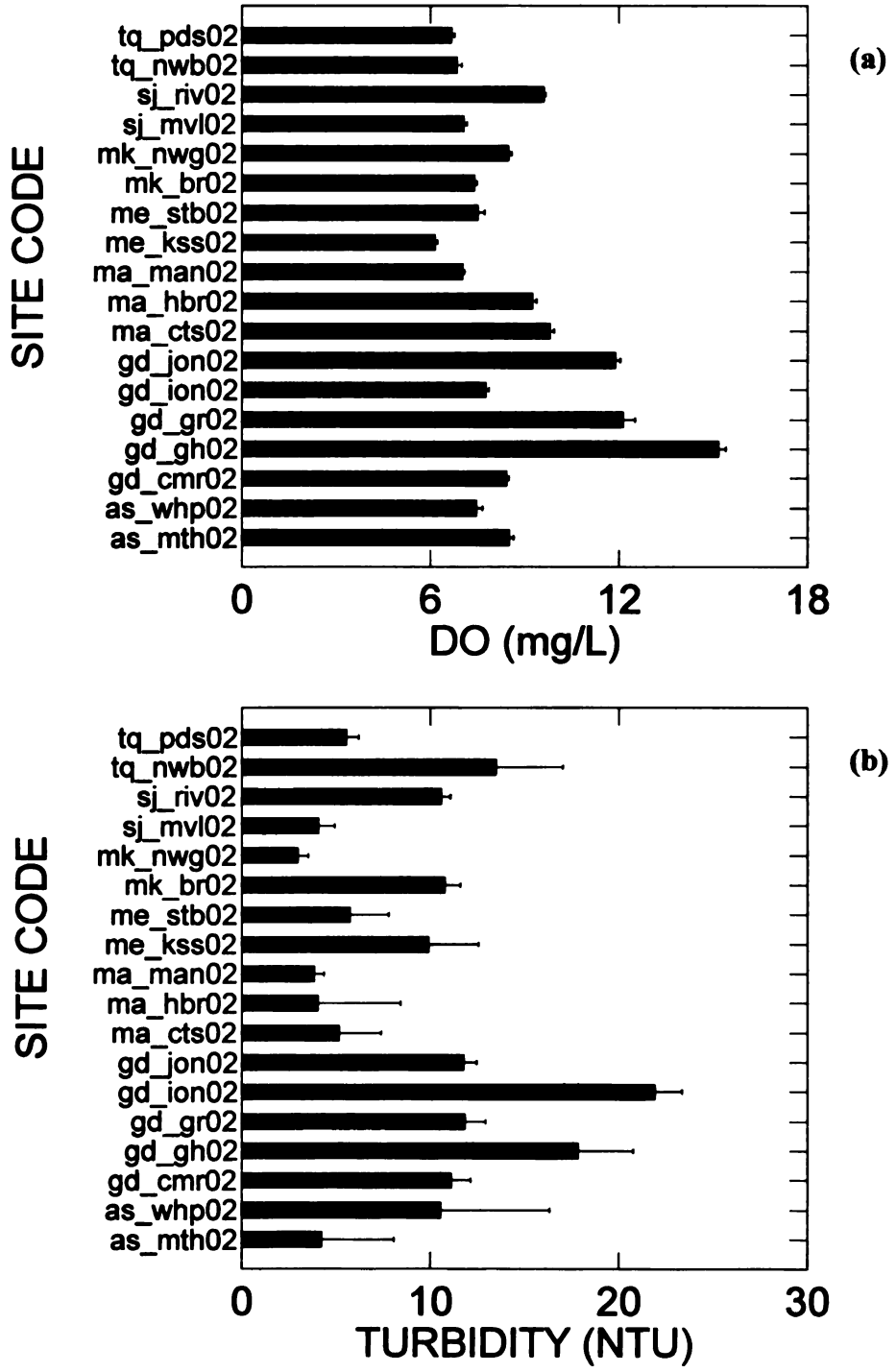

Figure 2.10. Mean (a) dissolved oxygen and (b) turbidity for all 2002 sites. Error bars indicate standard error. See Table 3.1 for site codes.

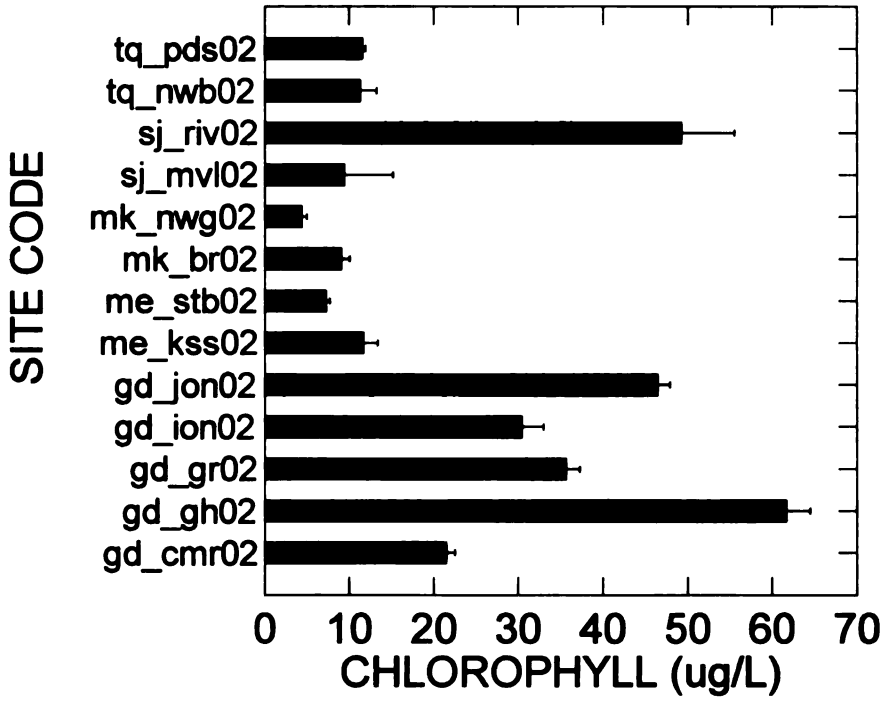

Figure 2.11. Mean chlorophyll values for all 2002 sites. Error bars indicate standard error. No chlorophyll data exist for the AuSable or the Manistee sites. See Table 3.1 for site codes.

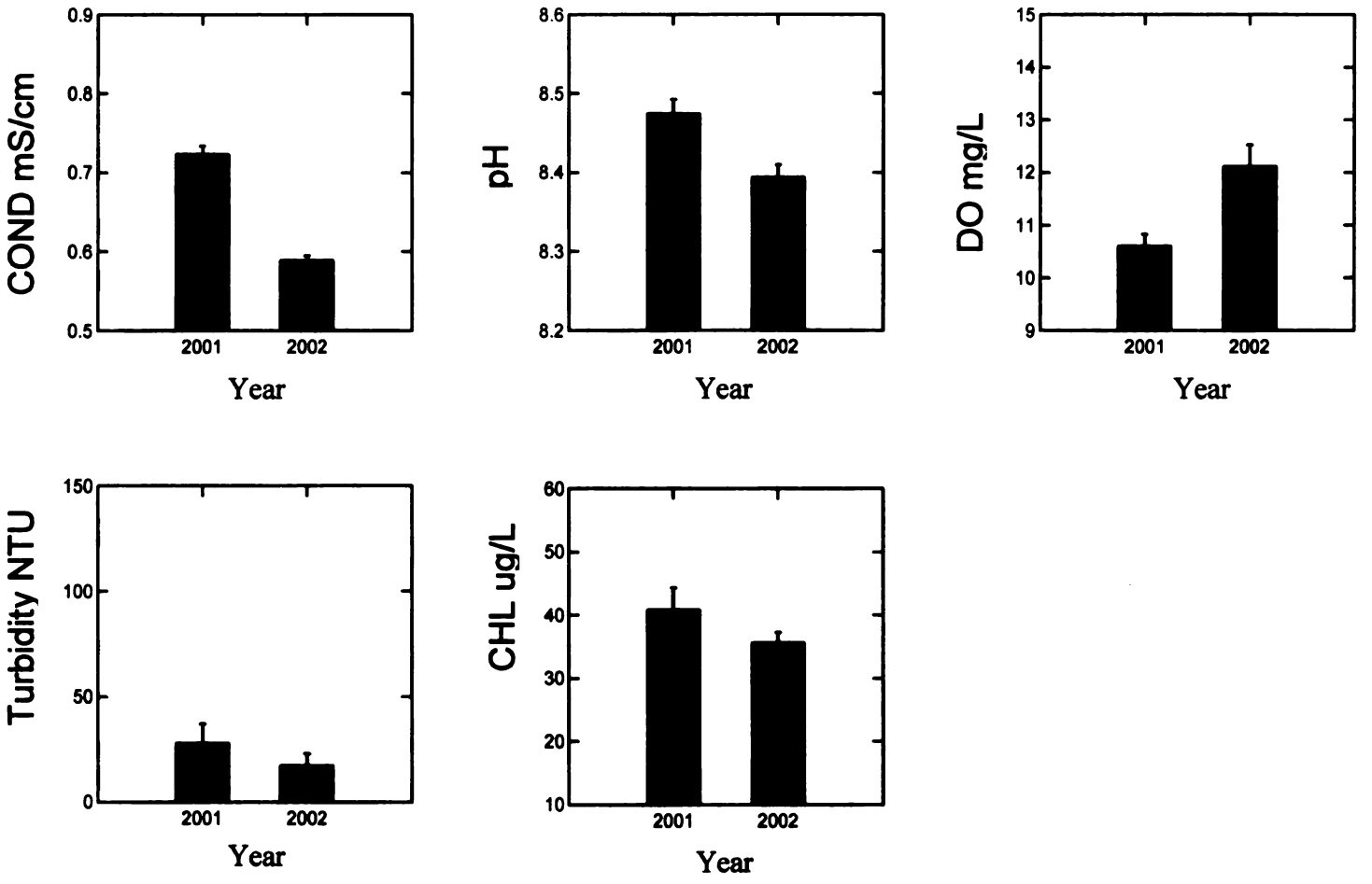

Figure 2.12. Comparison of 2001 and 2002 physical parameters from the Grand River @ Grand Rapids (gd_gr). All parameters were measured with a YSI 6600 multiparameter data sonde. Y-axis shows mean values +/- SE.

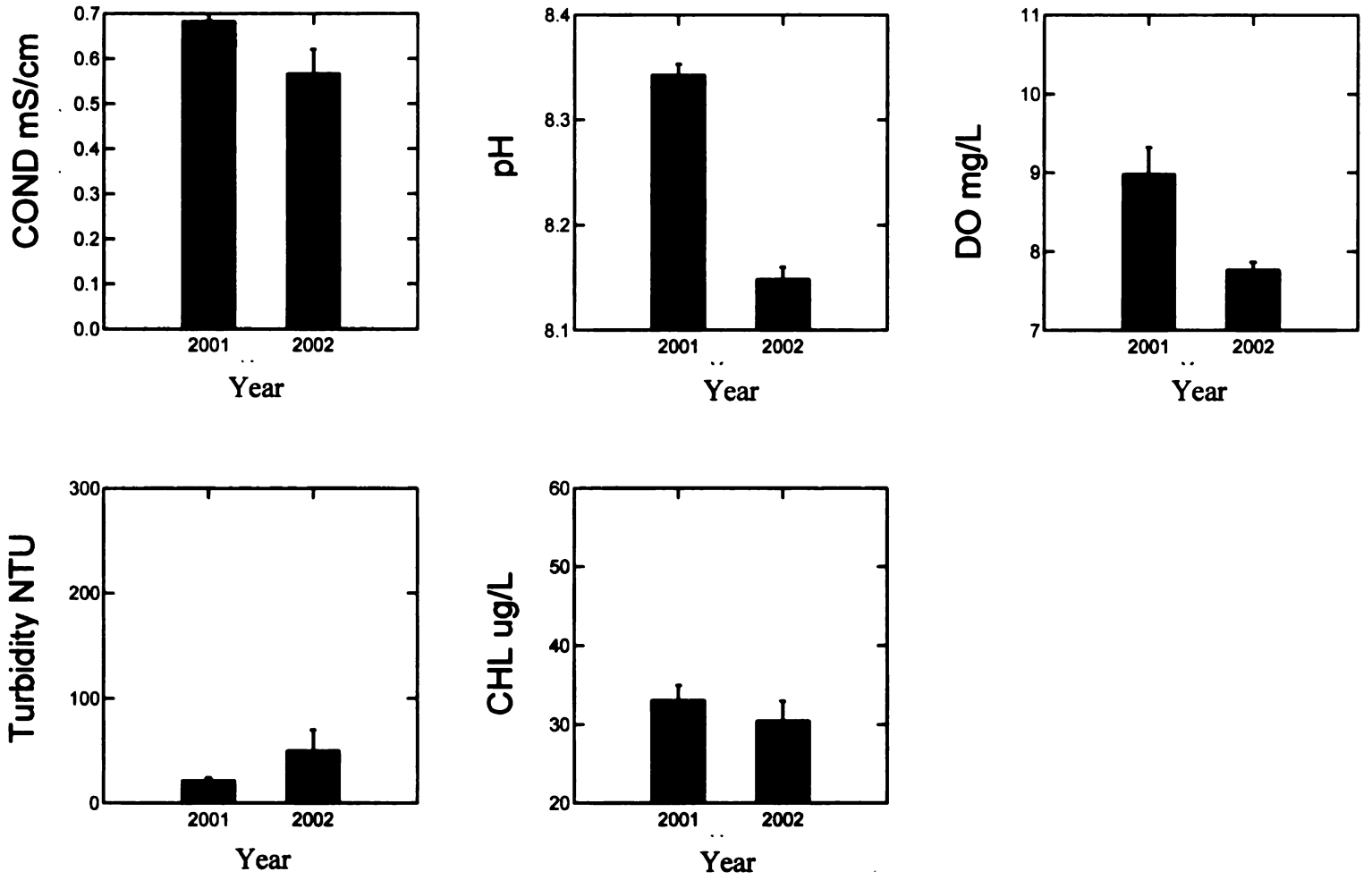

Figure 2.13. Comparison of 2001 ar ? physical parameters from the Grand River @ Ionia (gd_ion). All parameters were measured with a YSI 6600 multiparameter data sonde. Y-axis shows mean values +/- SE.

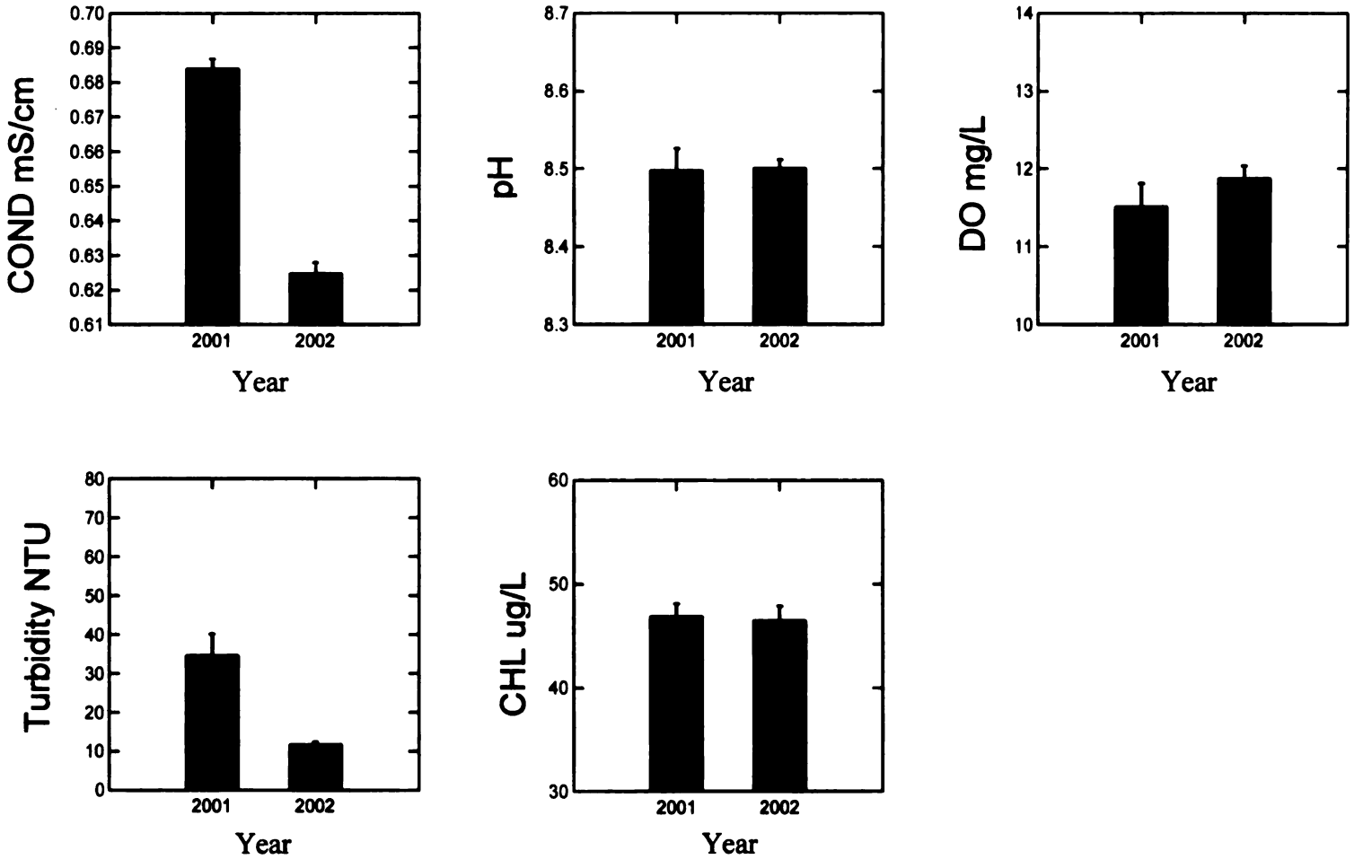

Figure 2.14. Comparison of 2001 and 2002 physical parameters from the Grand River @ Johnson Pk (gd_jon). All parameters were measured with a YSI 6600 multiparameter data sonde. Y-axis shows mean values +/- SE.

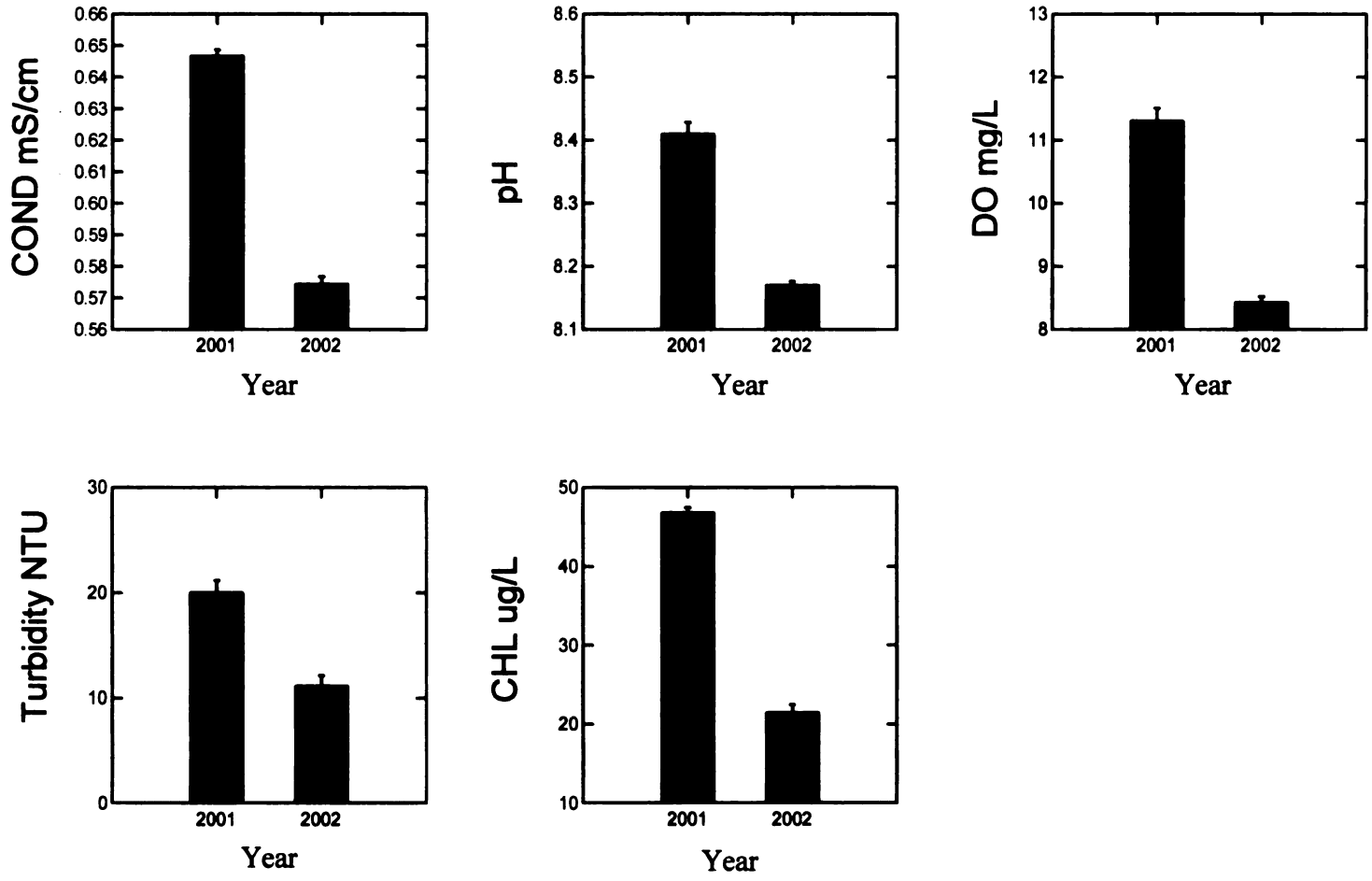

Figure 2.15. Comparison of 2001 and 2002 physical parameters from the Grand River @ Comstock Riverside (gd_cmr). All parameters were measured with a YSI 6600 multiparamter data sonde. Y-axis shows mean values +/- SE.

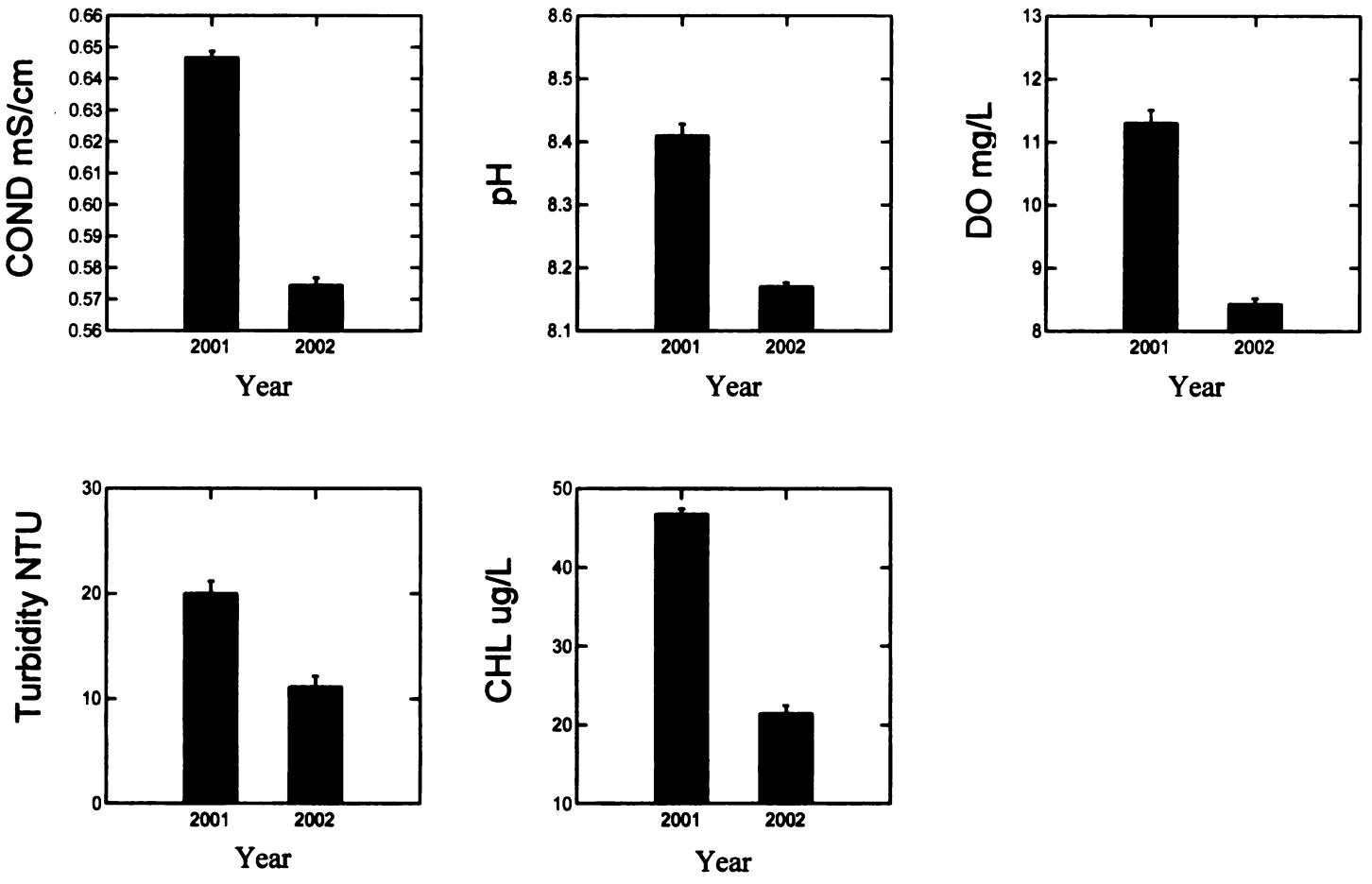

Figure 2.15. Comparison of 2001 and 2002 physical parameters from the Grand River @ Comstock Riverside (gd_cmr). All parameters were measured with a YSI 6600 multiparamter data sonde. Y-axis shows mean values +/- SE.

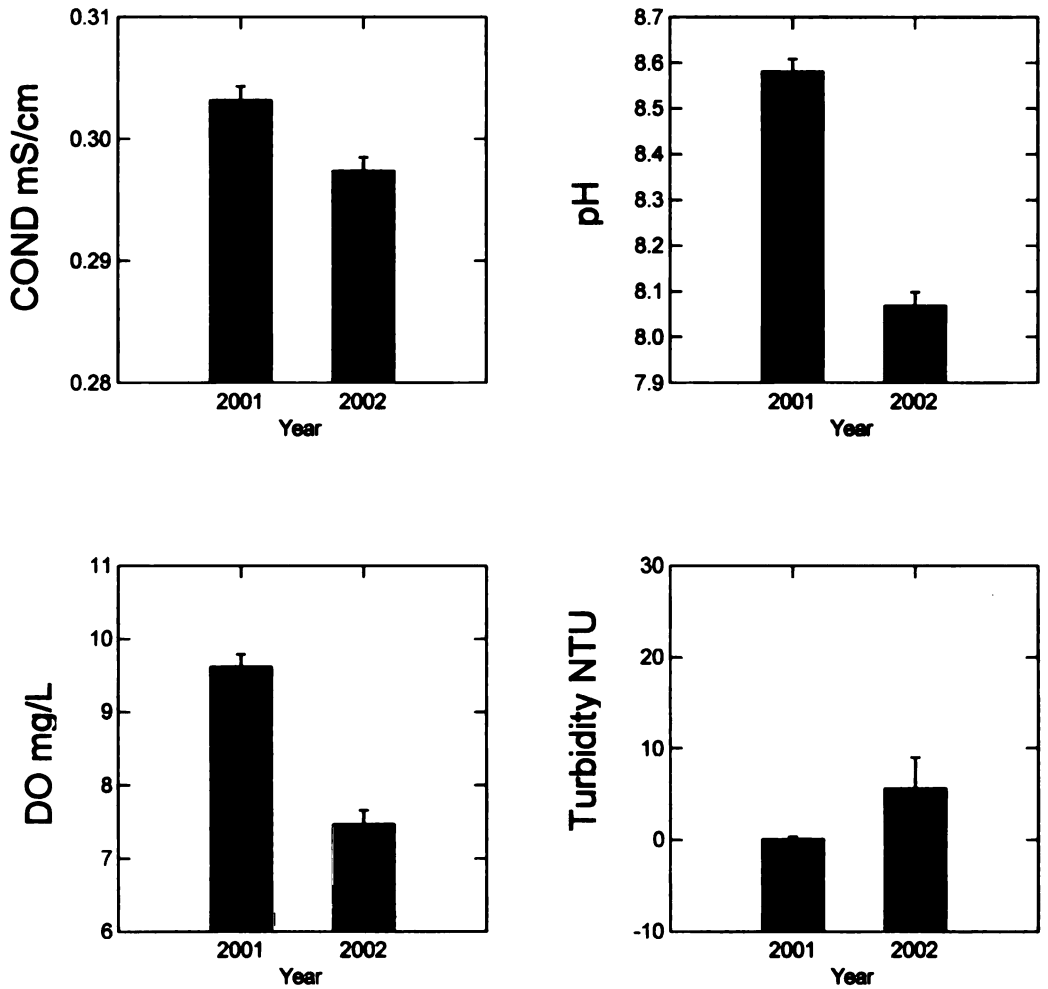

Figure 2.16. Comparison of 2001 and 2002 physical parameters from the AuSable River @ Whirlpool (as_whp). All parameters were measured with a YSI 6600 multiparameter data sonde. Y-axis shows mean values +/- SE.

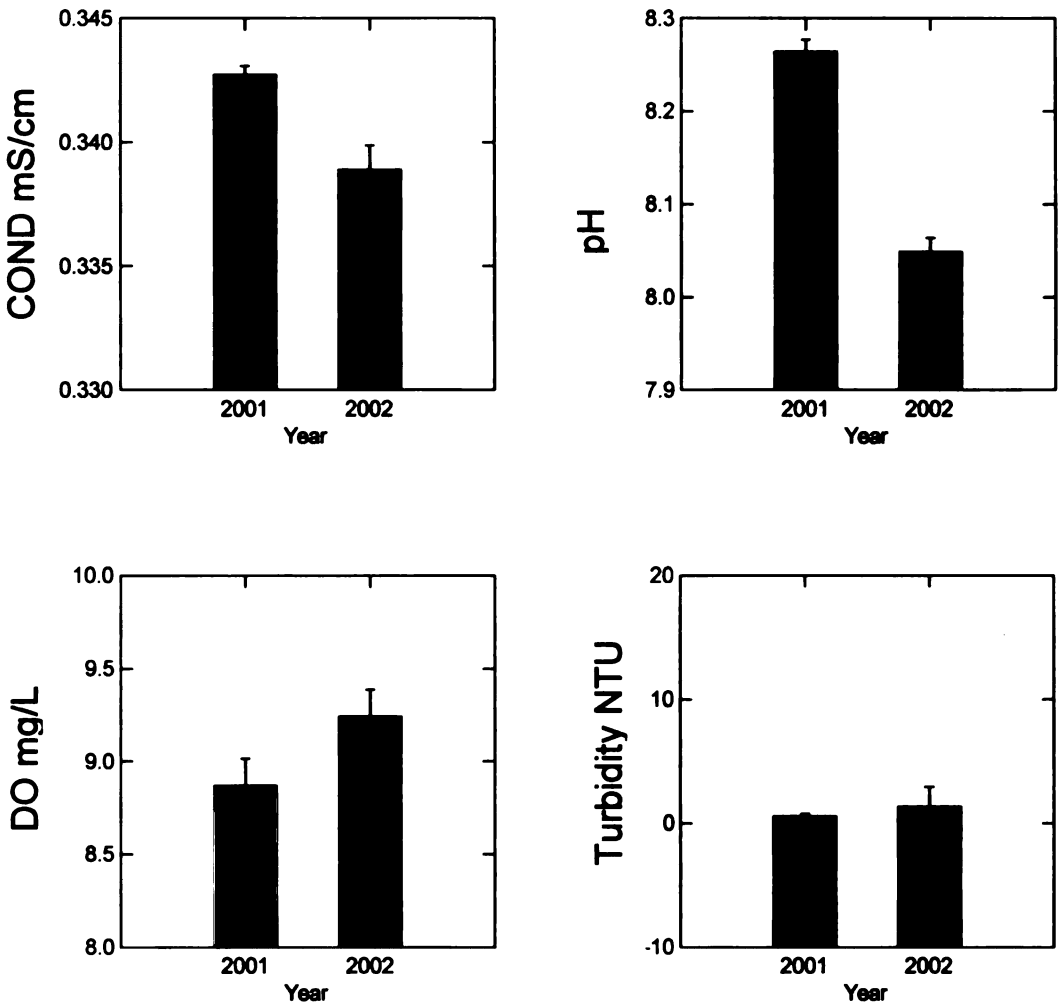

Figure 2.17. Comparison of 2001 and 2002 physical parameters from the Manistee @ High Bridge (ma_hbr). All parameters were measured with a YSI 6600 multiparamter data sonde. Y-axis shows mean values +/- SE.

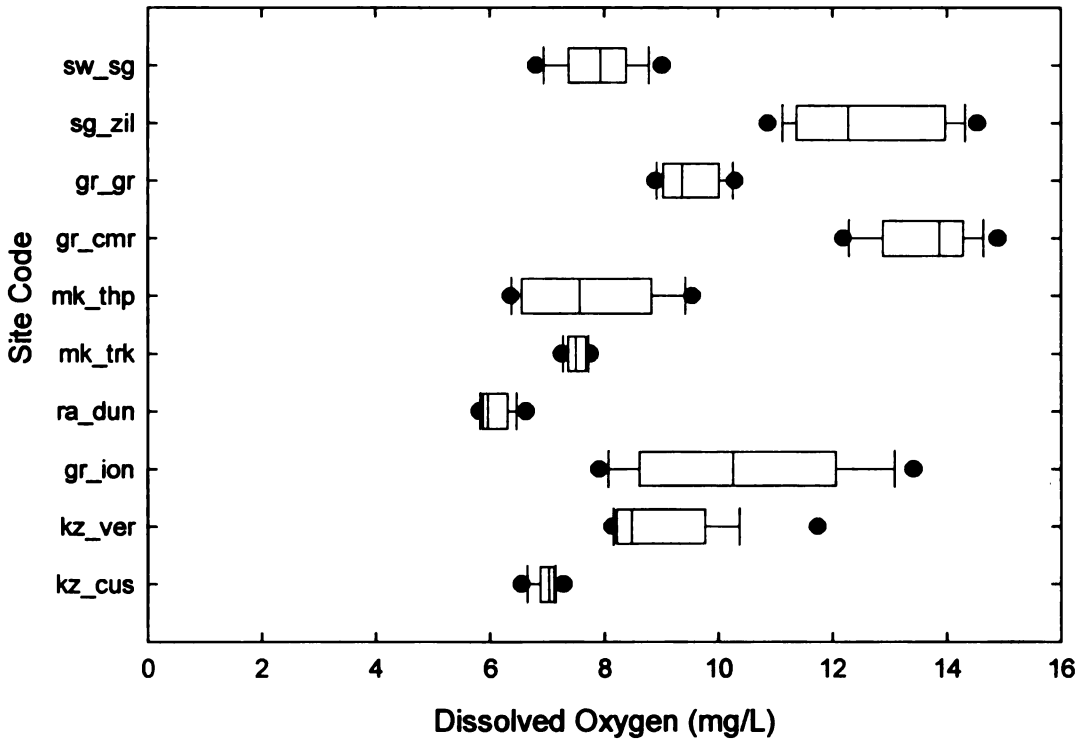

Figure 2.18. Box and whisker plot of diel oxygen change for selected 2001 sites. Data are from a YSI 600 multiparameter data logging sonde.

CHAPTER 3:

BIOLOGICAL EVALUATION OF MICHIGAN'S NON-WADEABLE RIVERS USING MACROINVERTEBRATES

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BIOLOGICAL EVALUATION OF MICHIGAN'S NON-WADEABLE RIVERS USING MACROINVERTEBRATES

INTRODUCTION

Large river ecosystems have long been subjected to problems caused by human settlement. Since the beginning of civilization, these systems have been relied-upon heavily for irrigation, navigation, waste removal, and drinking water, and have subsequently paid a heavy price for their utility to human inhabitants. Because of their large watersheds, large rivers are subjected to a much more complicated suite of problems than small streams. These include upstream and watershed influences from agriculture, logging, and urban development, introduction of exotic species from international shipping, as well as damming and dredging for navigational purposes. This has resulted in many of our large, non-wadeable rivers being severely degraded (Paton 1979; Sparks et al. 1990; Gore and Shields 1995; Sparks 1995).

Biological assessment has become an accepted way of evaluating the synergistic effects humans have on lotic environments (Cairns 1990; Cairns and Pratt 1993; Karr 1993; Karr 1987; Karr and Chu 1999; Kerans and Karr 1994). These protocols are preferable to chemical monitoring primarily because they integrate effects of short-term environmental variation and combined effects of water and habitat quality, as opposed to mere snapshot of water quality at the time when chemical samples are taken. Biological assemblages commonly used in such protocols include fish, macroinvertebrates, and algae, all with different levels of spatial and temporal resolution. Macroinvertebrates are

particularly useful because they are abundant in most streams, provide indication of localized (reach scale) conditions, are easy to collect and identify, and serve as a food base for higher trophic levels (Cairns and Pratt 1993).

The development of effective indicators to assess the ecological condition of large river ecosystems is becoming a priority for state and federal agencies (USEPA). However, most bioassessment protocols for macroinvertebrates have been developed almost exclusively for small, wadeable streams as a means to evaluate nutrient enrichment and oxygen depletion (e.g., MDNR 1991). Because of the fundamental differences between small stream and large river structure and function (e.g., Vannote et al. 1980), some metrics included in these protocols are not necessarily applicable to large rivers.

Our understanding of large river ecology has lagged compared to that of smaller, wadeable streams. Sampling difficulties relating to their depth, discharge, and structural complexity is a main reason for this. Additionally, the complicated factors which shape non-wadeable biological communities (watershed landuse, in-stream habitat, and water quality issues), make causal relationships between stressors and biota difficult to define using what is known from research on wadeable streams. However, the application of fundamental stream theories such as the River Continuum Concept (Vannote et al. 1980), the Serial Discontinuity Concept (Ward and Stanford 1983), and the Flood Pulse Concept (Junk et al. 1989) to large rivers have received attention (Bayley 1995). These fundamental theories have facilitated our understanding of large river ecosystems (Johnson et al. 1995), in that they have served as templates to set expectations of large

river structure and function, and have helped formulate ideas regarding the effects humans have on these systems (Johnson et al. 1995).

The need for ecosystem management of large river systems is extremely important (Sparks 1995). Currently, some governmental agencies are in the process of developing habitat and biological sampling protocols for non-wadeable river assessment (Lazorchak et al. 2000), and these protocols include methods for assessing macroinvertebrate communities (Klemm et al. 1999).

My primary objective was to develop a non-wadeable index of biological integrity (NW-IBI) using macroinvertebrate attributes that best describe variability in ecological condition of Michigan's non-wadeable rivers. I developed the protocol through a systematic approach of reducing variable redundancy, and determining which macroinvertebrate attributes were most responsible for among-site differences. Metrics selected for the final protocol describe population, community, and functional differences among sites. The goal was to develop two separate IBIs—one using composite samples from all habitats present in each study reach, and one using samples from targeted habitats.

When used in conjunction with the non-wadeable habitat index (NWHI; Wilhelm et al. 2005; Wilhelm 2002), the NW-IBI will provide an objective means of evaluating anthropogenic impact and targeting specific rivers and segments of rivers for conservation or restoration.

METHODS

Defining Non-wadeable Rivers

The first step in undertaking such a project is to define what 'non-wadeable' means. Intuitively, a non-wadeable river is either too deep or discharge is too high to permit one to safely wade into in order to acquire samples needed for biological (or habitat) assessment. However, this necessitates actual field visits, which is not necessarily cost-effective. For this reason, it is desirable to established basic guidelines to help us identify non-wadeable segments of rivers before going out in the field.

Large rivers have been defined in the literature in many ways, including those whose drainage basins exceed 1600 km^2 (Ohio EPA 1989), have an average depth of greater than one meter (Stalnaker et al. 1989), or a stream order of 6^{th} or higher (Sedell et al. 1989; Johnson et al. 1995; Vannote et al. 1980). For a more detailed discussion, see Wilhelm (2002).

I used USGS gauge data and various GIS layers to define and identify non-wadeable rivers of Michigan as those of order 5 and above, with drainage areas greater than 1600 km^2, mainstem lengths exceeding 100 km, and mean annual discharge greater than 15 m^3/s. These criteria usually translated to those rivers and river segments which are either too deep or discharge is too high to safely acquire samples without the use of a boat, and identify 22 rivers in Michigan with non-wadeable sections (Table 3.1).

Study Sites

Rivers visited ranged from 5^{th} to 7^{th} order and were subject to a range of human influences and natural variability. Through the course of this study, I sampled 33 non-

wadeable river reaches in 11 rivers in Michigan (Table 3.1). Several of these sites were repeated. I sampled sites in eleven major watersheds, ranging in size from 16,856 km2 (Saginaw River) to 2,124 km2 (Taquamenon River) (Table 3.1; Wilhelm 2002). Overall, sites were selected from the three major ecoregions in Michigan. Six watersheds were in the Southern Lower Peninsula (SLP), three in the Northern Lower Peninsula (NLP), and two were in the Upper Peninsula (UP) (Table 3.1). These ecoregions encompass a considerable range in climate, vegetation, geology, and human landuse. A discernible gradient in current and historical landuse exists from north to south in Michigan's watersheds. Natural areas dominate the UP (90% forested or covered by wetlands). Though logged in the late 19th century, most of the NLP today is dominated by mixed conifers and deciduous trees (75% natural; <4% urban, <11% agricultural). Our NLP sites were the most heavily influenced by humans—less than 25% remained as natural land, with 57% agricultural and over 8% urban (Wilhelm 2002).

Site Selection

When undertaking a project such as this, especially when no true reference condition exists, it is important to sample the full range of impact levels to obtain an accurate picture of what these impacts have on macroinvertebrate communities. In choosing sites, I wanted to sample the entire gamut of ecological conditions, from those most highly impacted by urban development and/or agricultural development to those that are relatively undisturbed.

Wilhelm (2002) rated sites based on percent agriculture and urban landuse in the basin and in a 100m riparian buffer, dam density, NPDES permit density, and road

density. These scores were summed and incorporated into an overall watershed-level Catchment Disturbance Gradient (CDG) (Table 3.2). Sites were also rated based on number of gaps in the riparian and riparian width using aerial photographs, and these scores were incorporated into a Riparian Disturbance Gradient (RDG) (Table 3.3).

Sites visited ranged widely in both scores (Wilhelm 2002). Of the sites visited by the biological crew, the Tahquamenon River at Paradise scored best for watershed-level disturbance (CDG=0), while the River Raison at Dundee scored the worst (CDG=13) (Table 3.2). The AuSable River at Whirlpool scored the best for Riparian disturbance (RDG=0), while the St. Joseph River at Riverview scored the worst (RDG=8) (Table 3.3). It should be noted that a reach with a low score for either of these measures of anthropogenic influence should not necessarily be considered a reference reach, rather, it should be considered to be the least impacted condition. Because of the size of non-wadeable river watersheds, virtually all large rivers in Michigan are impacted by human settlement or other landuse issues in some way.

Field Methods

Reaches were defined as a 2000m section of river in which 11 evenly spaced transects (every 200m) were sampled for habitat quality, water quality, and macroinvertebrates (Figure 3.1). U.S. EPA studies have found that, for Midwestern streams, a reach length of 40 channel widths is sufficient to characterize the variability in fish communities (Lazorchak et al. 2000). Despite the fact that the average wetted width of our sites was 89m (range=32-183m), I believed this to be a sufficient length in capturing most of the natural variability in macroinvertebrate communities (as well as

habitat) in each reach, while keeping the length of the study reach feasible for rapid protocols.

At each study reach, I used procedures modified from US EPA protocols for sampling non-wadeable rivers macroinvertebrates. I sampled all available habitats at each transect. Habitats were categorized into 6 different types: fine particulate organic matter (FPOM), sand, coarse substrates (gravel and small stones), cobble, large woody debris (LWD), and macrophytes. Each habitat was sampled with a D-frame aquatic dip net with 500 μm mesh in 15 second timed sweeps to standardize the sampling effort. Samples were then rinsed in a sieve (500 um) and preserved in the field with 70% EtOH.

Each reach was sampled twice. The first set of samples was kept separate by habitat to look at habitat-specific differences in macroinvertebrate community structure. The second set of samples was combined into one large composite sample for each site. This overall sampling scheme is a slight modification of the U.S. EPA protocol for sampling non-wadeable rivers (Lazorchak et al. 2000).

Samples were sorted and identified to family level in the laboratory. Habitat-specific samples were completely sorted, while the large composite samples were subsampled into quarters before being processed.

Many state and federal agencies target specific habitats to help control for differences in macroinvertebrate populations and communities due to differences in habitat. For example, cobble samples from two different rivers might be more similar (e.g., in terms of diversity or FFG composition) than a cobble and a sand sample from the same river. Another benefit of habitat-specific sampling is that it reduces the amount of fieldwork for assessment crews and can help reduce the amount of detritus in samples.

At each transect, I recorded the different habitat types present and kept one set of samples separate to determine whether or not habitat-specific sampling was possible for the biological assessment of non-wadeable rivers. This process was mainly one of observation. Which habitats were common to all of our study reaches? Is macroinvertebrate structure in any given habitat variable as a result of water quality or other human influences? How does habitat type affect metric robustness?

Data Reduction and Metric Selection

For each site, an array of different summary attributes, or potential metrics, were calculated. These potential metrics described population, community, and functional levels of organization. In total, 26 metrics were included in the initial analysis (Table 3.5). However, many of these metrics were highly correlated with each other, and some did not vary among sites. Because of this, we used a stepwise process in selecting metrics to include in the final protocol.

The first step was to eliminate redundancy among metrics. A Pearson correlation analysis was done on all potential metrics to see which ones were highly correlated. If two or more individual metrics were highly correlated (correlation coefficient > 0.70), metrics were removed from further analyses based on several criteria, precision of metric response to individual stressors, biological meaningfulness, ease of measurement, and distributional characteristics. Correlation analysis was done on population, community, and functional metrics separately to ensure that each type of metric was represented in the final protocol.

The second step in metric selection was to perform principal components analysis (PCA) on metrics retained from the correlation analysis. This provided information on which biological attributes were most responsible for among-site variation. Again, criteria were set for the retention of metrics from the PCA. Only axes with eigenvalues greater than 1 were further examined, and retained metrics were those with the highest loadings on each retained axis. For this part of the analysis, all metrics were standardized by mean and standard deviation because of vast differences in scale of individual metrics. When necessary, metrics were also transformed to approximate normal distributions.

Metric Response to Stressors

Examining metric response to human influences is a crucial part of the metric selection process. In the first two steps of the data reduction process, metrics were identified that provide unique information and explain significant among-site variation. However, if metric response to human influences is not log-linear or exponential, an evaluation of sites based on that metric will be ambiguous. For example, if a metric value is plotted for each site along a gradient of human influence and the response is quadratic, highly impacted sites and unimpacted sites may receive the same score (Stevenson and Smol 2001). The metrics examined (Table 3.5) are generally agreed to have predictable responses to human influences, with the exception of some of the FFG attributes (Ohio EPA 1989; Barbour et al. 1992).

To determine the specific stressors to which individual metrics respond, we used multiple linear regression (MLR) with individual metrics as the dependent variables and a suite of physical, chemical, habitat, and landuse variables as the independent variables.

Both forward and backward stepwise regression (tolerance=0.001, p to enter/remove=0.15) helped us compare environmental variables to which metrics respond. Significant metric/stressor relationships as well as overall explanatory power (R^2) helped us evaluate each regression model.

Environmental variables were recorded at each transect at each site, and mean values per site were used in the regression analysis. Physical and chemical variables (DO, temperature, pH, conductivity, turbidity, and suspended chlorophyll) were measured with a YSI® 6600 multiparameter sonde. Water samples were taken at 3 transects (A, G, K; Figure 3.1) per site for measuring nutrient (Total P, Total N) levels at each site. Landuse variables (% urban, agricultural, and natural) at the watershed scale and in a 100m riparian buffer were calculated using GIS data by Wilhelm (2002) and Wilhelm et al 2005.

These analyses were conducted separately for both the composite samples and the habitat-specific samples.

Evaluation of the NW-IBI

The NW-IBI was evaluated using several techniques. The first technique was to randomly choose a subset of sites within each assessment category ("poor", "fair", "good", "excellent") and designate them as model sites. I used discriminant function analysis (DFA) on the model sites with all metrics retained for the final protocol as descriptors. We then used these functions to assign the remaining test sites to assessment categories. We also performed DFA on all sites with jackknifing, where one site is removed from the list of sites in an iterative fashion to construct the overall model. In

both cases, DFA determines the percent of correct classifications, and this can be used as a means of evaluating the overall model's efficiency at classifying sites (Legendre and Legendre 1998).

This method of evaluation, while valuable, could be considered circular because I selected metrics using all of our study sites. We also evaluated the NW-IBI by plotting scores from each sites against mean site rankings. These rankings were based on the CDG, the RDG (Wilhelm 2002), as well as number of transects with large woody debris. Site rankings were calculated based on these factors individually, and then mean site ranking was determined (Table 3.24). By comparing these rankings to the NW-IBI scores for each site, the protocol is evaluated with independent expectations for each site.

In addition to evaluating the NW-IBI scoring system with independent expectations of site-specific ecological integrity, we also evaluated the NW-IBI's sensitivity to anthropogenic influences at different spatial scales (e.g., watershed vs. riparian vs. overall habitat). We used regression analysis to look at the relationships between the NW-IBI and the RDG, CDG, and the overall Habitat Index (HI) developed by Wilhelm (2002). This also helped use evaluate the composite and the habitat-specific assessment types' sensitivity to the collection of influences that may impact biological integrity.

RESULTS

Non-wadeable Macroinvertebrates

We found a wide variety of macroinvertebrates in our study reaches. Overall, there were a total of 17 non-insects, with gastropods accounting for the most non-insect

richness and crustaceans accounting for the highest non-insect abundance. There were a total of 76 insect families in all of our samples with Diptera, Ephemeroptera, and Trichoptera making up the majority of insects collected (Table 3.4).

Habitat-Specific Assessment

From our observations in the field, we noticed that the only habitat common to almost all of our study reaches was fine particulate organic matter (FPOM), and there was very little difference between macroinvertebrate diversity or taxa richness in FPOM samples taken from a highly impacted river and FPOM samples from rivers that were fairly unimpacted (Figure 3.2). Cobble habitats were extremely rare in most sites, and macrophyte habitats varied seasonally. Sand and coarse substrates varied more by watershed, and would presumably be more influenced by high discharge events (personal observations). Because large woody debris (LWD) habitats were usually fairly abundant in most of our reaches (2 sites contained no LWD: sw_sg and tq_nwb) (Table 3.1), we decided to use LWD as our target habitat for the habitat-specific assessment. Large woody debris has been shown to be an extremely important habitat for benthic macroinvertebrates (Hilderbrand et al. 1997; Abbe and Montgomery 1996), and has been shown to harbor the highest insect diversity in large rivers (Merritt et al. 1996). Samples taken from LWD indicated that taxa richness and diversity showed relatively little variance within reaches, yet varied significantly among rivers (Figure 3.2).

This enabled me to construct two types of biological assessments: one using composite data for all habitats present, and the other using only large woody debris (LWD) sample data. While a composite assessment is more time consuming due to the

increased amount of detritus compared to LWD samples, it may be necessary in rivers with insufficient amounts of LWD. The advantage to habitat-specific sampling is that, by focusing on a single habitat, water quality issues are less likely to be masked by the variation inherent in samples from different habitats (Parsons and Norris 1996). However, composite assessments may be the only option in some rivers with insufficient LWD habitat.

Data Reduction: Correlation Analysis

The correlation analysis resulted in a total of 9 metrics being discarded due to redundancy, and these results hold true for both the composite data and the LWD data unless otherwise noted. In the group of population-level metrics, E_RICH and T_RICH were both discarded because of the high correlation with EPT_RICH (Tables 3.6-3.7). Of the community-level metrics, PER_E and EPT_DIP were discarded due to their high correlation with PER_EPT, and DIV was discarded because of its high correlation with both RICH and PER_DOM. (Tables 3.8-3.9). PER_OLIG was retained for the composite sample analysis, but discarded for the LWD analysis due to very low numbers of Oligochaeta in the LWD samples (Table 3.5). The correlation analysis resulted in the elimination of P_R, C_FPOM, and T_BFPOM due to high correlations with SCR, SHD, and CF, respectively (Tables 3.10-3.11). The decision to retain or discard certain highly correlated metrics was based on ease of measurement and best professional judgment. For example, RICH and DIV were highly correlated with each other, but RICH is much easier to measure, so it was retained. See Table 3.5 for metric codes and a summary of metrics retained during the correlation analysis.

Data Reduction: Principal Components Analysis

Based on the criteria outlined above, the first 5 PCA axes were further examined for the composite samples (Figure 3.3). All other axes had eigenvalues less than 1 (Figure 3.3; Table 3.12). Because some metrics had similar loadings on individual axes, sometimes more than one metric was retained for the final protocol from any given axis. Overall, the PCA analysis for the composite metrics resulted in a total of 8 metrics retained (Table 3.13). From axis 1, two functional metrics were retained (the Habitat Stability FFG surrogate and FFG Diversity). Only Percent Trichoptera was retained from axis 2. The population level metric, EPT Richness, and the community level metric, Taxa Richness, were both retained from axis 3. Plecoptera Richness and Diptera Richness were both retained from axis 4, while only Percent Dominance was retained from axis 5 (Table 3.13). See Table 3.5 for a summary of metrics retained in the PCA analysis.

Because all metrics did not describe an equal amount of among-site variation, each was weighted based on the axis from which it was retained. Both the habitat stability FFG surrogate and FFG diversity were retained from the first PC axis. This axis described 50% of the overall variation among sites, and so each metric is based on a 25-point scale. Likewise, % Trichoptera abundance is based on a 20 point scale, EPT taxa richness an 8 point scale, total taxa richness a 7 point scale, and the remainder are based on a 5 point scale. The total possible score for the composite NW-IBI is 100 points (Table 3.15).

Principal components analysis resulted in the further examination of axes 1-4 (Figure 3.3; Table 3.12), and a total of 6 metrics were retained for the final protocol for the LWD assessment (Table 3.14). Percent Dominance, Habitat Stability FFG surrogate, and FFG Diversity were all retained from axis 1. Percent Trichoptera, Diptera Richness, and percent Scrapers were retained from axes 2, 3, and 4, respectively, based on their principal component loadings (Tables 3.14-3.17).

As in the composite assessments, individual metrics should have weighted contributions to the overall LWD NW-IBI based on the axis from which it was retained and that axis' corresponding contribution to the overall variation among sites. Axis 1 described approximately 60% of the overall variation, and since 3 metrics (HAB_STAB, FFG_DIV, and PER_DOM) were retained from this axis, each are scored on a 20 point scale. Percent Trichoptera and Diptera taxa richness are both scored on a 15 point scale, and % scraper abundance is scored on a 10 point scale. Like the composite assessment, the total possible score for the LWD NW-IBI is 100 points (Tables 3.16-3.18).

Overall, sites showed no real tendency to cluster based on river basin (e.g., Figure 3.4); however, they showed a slight tendency to cluster based on ecoregion on axis 1 (Figure 3.5a). This is not surprising, due to the condition of sights in the northern ecoregions (NLP and UP). ANOVA showed an overall difference in RDG ($F=5.560$, $df=2$, $p=0.009$) and CDG ($F=45.332$, $df=2$, $p<0.001$) scores by ecoregion Fisher's LSD pairwise test showed significantly higher RDG as well as CDG scores for SLP compared to UP sites (Figure 3.6). This overall pattern of sites clustering by ecoregion was not observed on subsequent axes (e.g., Figure 3.5b).

Metrics Included in the Final Protocol and Stressor-Response Relationships

Defining metric response to stressors is key to effectively managing aquatic resources. Ideally, individual metrics are responsive to a particular stressor, which is caused by a particular human behavior. Macroinvertebrates in large rivers are subjected to a suite of stressors, ranging from water chemistry issues (e.g., pH, nutrients), instream habitat, riparian landuse, and watershed landuse. MLR helped identify specific stressors-response relationships, and overall, these relationships were relatively weaker for composite metric-stressor relationships (mean R^2=0.47) than LWD metric-stressor relationships (mean R^2=0.64).

Composite Assessment Metrics

Overall, metrics included in the final composite assessment protocol responded to a wide variety of human influences on several different spatial scales, from watershed landuse to instream habitat and water chemistry and instream habitat. Several metrics showed significant negative correlation with percent urban (e.g., HAB_STAB, RICH, EPT_RICH) and agricultural (e.g., HAB_STAB) landuse in the watershed. Other metrics showed significant correlations with percent natural landuse in the watershed (e.g., HAB_STAB) and in the riparian zone (e.g., PER_DOM). Percent Trichoptera showed significant correlations with agricultural landuse in the riparian zone. Both EPT_RICH and P_RICH had significant correlations with instream large woody debris.

Some metrics also correlated with various water quality parameters such as suspended chlorophyll (FFG_DIV, DIP_RICH), turbidity (FFG_DIV, DIP_RICH), conductivity (HAB_STAB, FFG_DIV, PER_DOM), and pH (DIP_RICH). Metrics also

responded significantly to total phosphorous (FFG_DIV, PER_DOM) as well as total nitrogen (DIP_RICH). For a detailed of composite metrics' response to specific stressors, see Table 3.19 and Appendix 1.

LWD Assessment Metrics

LWD metrics also showed a wide range of factors to which they respond, and it should be noted that the same metric sometimes showed different responses to stressors in the LWD assessment compared to the composite style assessment (Table 3.20). Some were significantly correlated with watershed landuse (e.g., HAB_STAB, PER_DOM, SCR) and riparian landuse (e.g., PER_DOM, PER_T).

Several LWD metrics were significantly related to water quality parameters such as pH (FFG_DIV, SCR), dissolved oxygen (PER_DOM), and conductivity (SCR). Like the composite metrics, some LWD metrics were also significantly correlated with total nitrogen (HAB_STAB, FFG_DIV, DIP_RICH) and total phosphorous (FFG_DIV, SCR). For a more details regarding LWD metrics' response to specific stressors, see Table 3.20 and Appendix 2.

Integration of Biological Metrics: The Non-wadeable Biotic Index (NW-IBI)

I set the scoring criteria to correspond to quartiles of each metric in a 4 bin scoring system by combining all our sites and calculating the 25th, 50th, and 75th percentiles for each metric. For example, for a metric scored on a 25 point scale (e.g., HAB_STAB in the composite assessment), sites scoring below the 25th percentile received a score of 0 for that metric. Sites scoring between the 25th and 50th percentiles

received a score of 8. Sites scoring between the 50th and 75th percentile received a score of 16, and sites scoring above the 75th percentile received the full 25 points for that individual metric (Figure 3.7). This was done for all metrics for both the composite and the LWD assessment types (Tables 3.17-3.18).

The Non-wadeable Index of Biological Integrity (NW-IBI) has a total possible score of 100 for both composite and LWD assessments. A total score between 0-25 results in a classification of 'poor' for that site; a site with a total score ranging between 26-50 is classified as 'fair'; a site with a total score between 51-75 is classified as 'good'; and a site with a total score between 76-100 is classified as 'excellent' (Tables 3.14-3.15).

When the NW-IBI was calculated for all sites, results differed slightly for composite vs. LWD type assessments. Using the composite method, there were 3 sites classified as excellent, 11 sites classified as good, 12 sites classified as fair, and 8 sties classified as poor. Using the LWD samples only, 5 sites were classified as excellent, 10 sites were classified as good, 8 were classified as fair, and 9 as poor (Figure 3.8). Overall, LWD assessments had more sites classified as poor and excellent compared to composite methods. Interestingly, however, both types of assessments usually classified sites similarly in terms of overall ranking (Figures 3.9-3.11).

Evaluation of the NW-IBI

Discriminant function analysis (DFA) was used to evaluate the robustness and sensitivity of both forms of the NW-IBI. When the overall DFA model was calculated based on a subset of model sites from each classification type (e.g., "poor", "fair",

"good", "excellent") based on the composite assessment methods, 75% of the test sites were correctly classified as poor, 33% were correctly classified as fair, 67% were correctly classified as good, and 100% were correctly classified as excellent (Table 3.23). When jackknifing was used, 88% of the sites were correctly classified as poor, 50% were correctly classified as fair, 55% were correctly classified as good, and 67% were correctly classified as excellent.

When DFA was performed based on the LWD assessments using a subset of (model) sites to compute the model, 100% of the sites were correctly classified as poor, 25% were correctly classified as fair, 100% were correctly classified as good, and 33% of test sites were correctly classified as excellent. When jackknifing was used, 78% of sites were correctly classified as poor, 75% as fair, 60% as good, and 50% as excellent (Table 3.23).

When evaluated using independent expectations for site condition (e.g., Mean Rank), composite assessment scores (Figure 3.12) showed a slightly weaker relationship to site ranking compared to the LWD assessment scores (Figure 3.13). Both assessment methods showed significant correlations with the CDG and the Habitat Index (HI), although the correlation was lower with the composite assessments (Table 3.25) than the LWD assessment (Table 3.26). In both cases, there was a higher correlation between the biological community scores and the HI (Tables 3.25-3.26).

DISCUSSION

General Discussion of the NW-IBI

Throughout the development of this protocol, we sampled a wide variety of non-wadeable sites throughout Michigan. These sites were subjected to a variety of anthropogenic influences, and ranged from reaches that were considered to be in excellent condition to those in dire need of mitigation. This is extremely important, since no real reference condition existed for rivers of this size in Michigan due to their large watershed size and concomitant human influences. These influences invariably affect a river's biological constituents (Karr 1997).

Both the composite and the LWD assessment protocols are designed to be independent of river basin. PCA shows us that site scores do not seem to cluster by river basin. In other words, metrics do not simply identify watersheds. Each study reach has a unique signature, and that signature depends on local processes (e.g., anthropogenic impacts associated with that particular reach) (Figure 3.4). The NW-IBI was also designed to apply widely throughout the state, independent of ecoregion. While overall site scores are higher along PC axis 1 (defining functional differences) (Figure 3.5a), this difference is not seen in subsequent axes (Figure 3.5b). As mentioned earlier, CDG and RDG scores are significantly lower in the SLP ecoregion compared to the NLP and UP regions (Figure 3.6). This suggests that NW-IBI scores should naturally be higher Northern Michigan. Because many of the stressors identified by MLR as having significant correlations with the two functional metrics (FFG_DIV and HAB_STAB) are directly correlated with the RDG (e.g., riparian landuse) and the CDG (e.g., watershed landuse), I believe that the differences among sites were due to differences in actual

ecological condition that are results of, at least in part, anthropogenic influences that go beyond ecoregional differences.

It was my goal to construct a protocol that helped discern human impacts on non-wadeable rivers in a hierarchical manner, incorporating metrics that describe ecosystem or functional attributes, community composition, and population-level changes. These different levels of resolution are subject to different spatial and temporal scales of impacts (Noss 1990). In both forms of the NW-IBI, functional and community level metrics were weighted higher than population level metrics (Tables 3.17-3.18). Although this is a result of the principal component analysis, it makes sense from an ecological standpoint. It is generally agreed upon that higher levels of organization (i.e., functional or community level) are more reliable than lower levels (i.e., population level) because of the high degree of background variation to which these lower levels are subjected (Cottingham and Carpenter 1998).

The Habitat Stability FFG surrogate (Merritt et al. 2002) and FFG Diversity both provide ways to evaluate the functional, or ecosystem-level, differences among non-wadeable rivers. While some of this difference is expected to occur naturally (Vannote et al. 1980; Cummins 1988), human landuse practices and their associate stressors have been shown to affect functional composition of riverine systems (Merritt et al. 2002; Cummins 1993; Cummins et al. 1989). The fact that these functional metrics retained from the first principal component axes and were included in both types of assessments suggests that the overriding differences among sites (and so the overall effect of stressors) results in primary functional degradation.

Metrics included in the final protocol also describe differences among non-wadeable macroinvertebrates at the community level. Total Taxa Richness, Percent Dominance and Percent Trichoptera have been shown to vary predictably with human influence (Ohio EPA 1989; Barbour et al. 1992). High macroinvertebrate taxa richness generally indicates functional redundancy (e.g., many different scrapers, shredders, etc.) a diverse food base for fish and other vertebrate predators. Percent dominance is highly correlated with diversity (Tables 3.8-3.9). Often, if one particular species or group is particularly dominant in an ecological community, it is because that group is especially tolerant to the stressor to which the system is subjected (e.g., low dissolved oxygen or high turbidity). This idea of tolerance is particular to certain groups of macroinvertebrates. Percent Trichoptera individuals reflect the overall composition of the community comprised by caddisflies. Caddisflies are generally considered a group that is intolerant to low DO levels (Hilsenhoff 1988;Jacob and Walther 1981). Many of the families that dominate non-wadeable rivers in Michigan are also dependant upon firm substrates like cobble or LWD for attachment (e.g., Brachycentridae and Hydopsychidae), low turbidity (so as not to clog nets), and are reliant upon healthy upstream processing of coarse organic matter as a food base (Vannote et al. 1980).

The number of Ephemeroptera, Plecoptera, and Trichoptera (EPT) families describes population level differences among sites, and is a metric in many IBIs (Karr 1987; Kerans and Karr 1994; Jacob and Walther 1981; Hayashi 1989; Cairns 1990; Barbour et al. 1992; Marshall 1993; 1993; Pinel-Alloul et al. 1996; [MDNR] Michigan Department of Natural Resources 1991). The number of EPT families has been shown to decrease due to nutrient enrichment from agricultural landuse (resulting in low DO), and

turbidity. They are also considered desirable because of their prevalence in the drift, providing certain fish species with an adequate food supply. Many EPT families, especially Plecoptera families, require clean, firm substrate free of sedimentation because of trophic (e.g., scraper mayflies) or respiratory constraints. Diptera Richness is another population level metric, which is included in both protocols, has a negative relationship with anthropogenic impacts. In highly-impacted streams, often the only Diptera group present is Chironomidae, which are known overall to be highly tolerant to low DO levels (Hilsenhoff 1988), which is often related to intensive agricultural landuse and sedimentation of fine organic matter. In systems without these influences, Diptera Richness tends to be higher, and often several functional groups are represented by the Diptera community (Table 3.4).

It is clear that individually, these metrics describe ecologically meaningful differences among sites based on functional, community, and population level attributes. When taken together as a single index, the NW-IBI, they will help describe the ecological condition of non-wadeable rivers at the reach scale. For example, the lowest scoring site scored with composite metrics, Saginaw River @ Zilwaukee (sg_zil01, NW-IBI composite score=0) (Figure 3.9), is subjected to a variety of human influences, and scored zero for all metrics. This reach of the Saginaw River is used extensively for shipping, is dredged, and its riparian vegetation is almost completely altered (Figure 3.14). This site also scored "poor" in the LWD assessment (Figure 3.10).

An example of a site scoring "fair" in the composite assessment is the Grand River @ Ionia (Figure 3.9). At first glance, this site looks like it's in good shape, but upon further examination, one sees that the riparian buffer, is very narrow (Figure 3.15),

with intensive agriculture dominating the overall landuse for the watershed (AgWS=63%; Table 3.20). This is an example of a site with "fair" scores in almost every individual metric, suggesting a moderate amount of human impact at all spatial scales. Interestingly, the LWD assessment classified this site as "poor" in both years. This is likely due to the heavy siltation on much of the LWD habitat, which would cause the LWD NW-IBI score to be much lower than the composite score, which masks some of the water quality issues because it is dependent on overall habitat quality.

The Manistee River @ High Bridge scored "good" in both 2001 and 2002 when scored with the composite method (Figure 3.9). This site is characterized by an intact riparian zone, clear water, and a variety of macroinvertebrate habitat, including a large amount of LWD (Figure 3.16). This particular site scored low on the habitat stability FFG metric, and this is likely due to embeddedness of its normally coarse and sandy substrate (Figure 3.16). In 2001, this site also scored "good" in the LWD assessment, but in 2002, it scored "excellent" (Figure 3.10). This suggests that LWD habitat improved in some way in the second year this site was evaluated. In the summer of 2002, flows were relatively higher because of much more precipitation (personal observation), which could have acted to scour the LWD habitat of its fine layer of sediment, subsequently making this habitat more favorable to a variety of macroinvertebrate groups. This is reflected mainly in a higher score for the habitat stability FFG surrogate (Figure 3.10). Alternatively, the LWD assessment is simply more sensitive to water quality parameters, and these parameters vary to a greater degree temporally than landuse or habitat parameters.

There were only three sites classified as "excellent" by the composite assessment. The site that scored the highest was the Manistee River @ Coates Rd (Figure 3.9). This study site scored the highest possible for almost every individual metric in the composite assessment. In general, the Manistee River is a fairly unimpacted river, but this particular site had exceptional habitat quality (macrophytes, LWD, and coarse substrates with very little FPOM). Overall, this site had much hydrologic variability, with slow-moving areas dominated by macrophyte beds, as well as deeper, faster areas with cobble and clean LWD (Figure 3.17). This site also scored "excellent" in the LWD assessment (Figure 3.10).

In general, the failure of the NW-IBI to classify one site similarly from year to year or by composite versus LWD can be helpful when diagnosing ecological condition or a particular study reach. For example, incongruities between composite assessments from year to year may help diagnose effects of drought (low flows). Inconsistencies between composite and LWD assessments in the same year (especially when combined with the HI) may help diagnose problems with specific habitats (e.g., embeddedness). However, the variation in scores may simply be a result of the great degree of natural variation in these systems, which is a primary reason an IBI for non-wadeable rivers is such a challenge. Alternatively, these incongruities associated with variation in NW-IBI scores from one year to another could simply be a result of real differences in water quality parameters from year-to-year (see Chapter 2).

Metric Response to Stressors

The technique I used to discern relationships between metrics and the stressors to which they respond needs further work. While the technique of establishing metric-stressor relationships after metric selection is often used, this could presumably result in losing some precision in the overall NW-IBI as well as with individual metrics. This is because metrics that were not selected for the overall index may have had higher overall correlations with individual stressors. For example, in a correlation analysis, percent Chironomidae had a the highest correlation of any of the original 26 biological attributes with urban landuse in the watershed. Metrics with similarly high correlations with this landuse measure include FFG_DIV, but it could be argued that the methods I used resulted in the loss of some precision. However, the results of PCA did not show significant variation among-site variation in metrics that had high correlations with any single stressor or human behavior, and this could be due to the complex interaction among multiple stressors and overall river quality. This is an issue that needs further examination.

Using regression techniques to quantify metric-stressor relationships, while encouraged (U.S.EPA 2000), should be done with caution. Aside from defining correlations instead of actual causes, there are other pitfalls in the way that we examined relationships with metrics and environmental parameters. First of all, the suite of possible stressors is far from complete (Tables 2.1-2.2). For example, we made no measure of sediment toxicity (metals, PCBs, pesticides/herbicides), and this is commonly a problem in large rivers, especially those draining urban watersheds (Smith et al. 1987; Young and Huryn 1999). Also, it is important to note the difference between stressors

and the human activities that influence the stressor. For example, a common stressor in lotic ecosystems is low DO. In order to manage this stressor, the human activities causing it must be identified, and in the case of DO, these activities may include clearing riparian buffer strips in agricultural areas, thus allowing nutrient enrichment of the waterway. While we included both landuse practices as well as the stressors caused by those practices in the MLR models for both types of metrics, technically, this is not appropriate.

Despite this, including the activities (e.g., landuse) that cause stressors to increase (e.g., water quality parameters) allows us to evaluate metric sensitivity to scale. It provides us insight into the many factors influencing macroinvertebrates in large rivers. Very few of the metrics in either protocol have only the actual stressor included in its MLR model—they also contain some sort of measure of landuse (Tables 3.19-3.20). This is likely because of the many ways a particular landuse affects different stressor levels. For example, a high percentage of urban landuse in the riparian zone would cause a decrease in riparian filtering capacity, which would cause elevated turbidity from road run-off (also causing deposition of other pollutants), decreased LWD habitat, increased embeddedness, and may even increase suspended chlorophyll by reducing shading (although this would be less of a factor in large rivers). Intensive agricultural landuse, whether in the riparian zone or the entire watershed, could cause increased nutrient levels, turbidity, and pesticide run-off, resulting in reduced levels of DO, among other things.

In summary, non-wadeable rivers are subjected to a wide range of stressors. Some of these originate upstream, some are a product of watershed-scale activities, some

are caused by riparian landuse practices, and some are a result of channel modification (e.g., dredging and impoundment). To compound this, human activities at different scales may result in the same type of stressors, and macroinvertebrates integrate the effects of these stressors regardless of scale. For these reasons, it is extremely difficult to establish extremely strong relationships between any one stressor and metric values in non-wadeable rivers. Inferring stressor-response relationships in large rivers should be undertaken case-by-case, using the guidelines in Appendix 1-2, but relying heavily on professional judgment.

Evaluation of the NW-IBI

When dealing with variable systems such as non-wadeable rivers, it is important to be aware of where this variation arises, what it means, and how each type of assessment can be affected by it. The discriminant function analysis (DFA) can tell us a little about each assessment type's ability to classify sites consistently. For the test sites only, DFA does a better job of classifying extremely poor sites and good (LWD) to excellent (composite) sites (Table 3.23). In the jackknifed DFA, % correct classification is increased overall, again suggesting the metrics chosen do a reasonably good job at detecting among-site differences (Table 3.23). However, the cut-offs in scores for each classification type are somewhat arbitrary—a site scoring 49 is classified as "fair", while a site scoring 51 is classified as "good". Some of the misclassification in both DFA models is likely due to this factor. These categories, while useful as narrative descriptions of overall ecological integrity, should nonetheless be used with caution.

Regression analysis with NW-IBI scores and Mean Rank also show a general trend that is expected—as a site's overall ranking decreases, so does its NW-IBI score (Figures 3.11-3.12). The low correlation coefficients for both assessment types, while initially troubling, should be expected because the factors used to compute Mean Rank (e.g., landuse and large woody debris), while useful for setting independent expectations for sites, summarize a relatively small amount of expected variability among sites. For example, no water quality parameters or substrate composition data were included in the calculation of Mean Rank.

As far as the NW-IBI's sensitivity to scale-specific factors, both types of assessment showed significant correlations with the CDG, Mean Rank, as well as the Habitat Index (HI) (Wilhelm 2002). The correlation with the HI was the highest (Tables 3.25-3.26). This was not surprising, because the HI incorporates differences among sites at many different spatial scales, from the entire watershed to the littoral fringe habitats sampled for macroinvertebrates. It is widely acknowledged that one of the main advantages to biomonitoring is that biological communities integrate synergistic effects of the multiple scales of influence that humans have on ecosystems e.g., (Fausch et al. 1984; Karr 1997; Rosenberg and Resh 1993). Aquatic organisms are affected by landuse within the entire watershed, local (riparian) landuse, upstream processes, water quality, and instream habitat quality.

Comparing Composite and LWD Methods of Assessment

The evaluation of both the composite and LWD protocols must additionally be addressed in terms of differences between each type of assessment: composite vs. LWD.

Upon immediate examination, it may seem preferable to simply conduct the LWD assessment. It is quicker due to shorter sample processing times, it seems to be more sensitive, and it is more independent of overall habitat quality. However, LWD assessments may not always be the way to proceed, and this depends largely on the overriding goal of the assessment. One must consider both the robustness and the sensitivity of each assessment method.

For example, the composite assessment was relatively more robust than the LWD assessment at correctly classifying sites into four broad categories (Table 3.23). However, when one compares this with the regression of NW-IBI scores and Mean Rank, we see that LWD assessments are more sensitive to differences among sites (Figures 3.11-3.12). The composite assessment seems to be more effective at incorporating overall variation among sites and consistently evaluating them, while the LWD assessment is more sensitive to variation, presumably because LWD harbors a more stable macroinvertebrate community in general. Put another way, the composite assessment is more robust to small changes in the environment, and the LWD assessment is more adept at detecting these changes.

This has wide ranging management implications. For instance, if the goal of the assessment is trend monitoring, the composite method may be preferable. Composite assessment tends to be more robust to overall change, and would probably not detect subtle changes in water quality that may occur on a relatively short temporal scale. However, if the goal is to compare two sites within a short time frame, LWD assessments may be more favorable because this type of assessment would be more sensitive to subtle differences between sites.

It should be noted that LWD assessments are not always possible because some large rivers in Michigan do not have sufficient LWD habitat to conduct such an assessment. Figure 3.18 shows the number of new taxa (families) acquired from each transect in a 2000m reach from a subset of LWD samples. After 8 transects were sampled, on average, no new taxa were collected (Figure 3.18). While this may not substantially affect some metrics (e.g., those involving relative abundance), there are clear implications for richness metrics (e.g., EPT Richness, Total Richness). Therefore, it is our recommendation that unless there is LWD habitat at no less than 8 of the 11 transects in a study reach (Figure 3.1), only composite assessments can be relied upon (Figure 3.19). Unfortunately, this means that only sites with at least 8 transects containing large woody debris can reliably assessed using the habitat specific methods, biasing the LWD assessment toward only relatively high-quality sites. General procedures for both types of assessment, along with materials needed to conduct the assessment, are summarized in Appendix 3.

CONCLUSION

While the approach outlined above provides general methods for the development of ecological assessments that may be applied to other systems and assemblages, it is important to note the limitations of this approach. The problem with developing a protocol using the methods outlined above lies with the identification of metric-stressor relationships. While there are many different approaches used to develop biological evaluations including the examination of rare species based on predictive models (Hawkins, Norris et al. 2000) and the use of functional feeding group surrogates for

ecosystem processes (Merritt et al. 2002), recent research points to the importance of developing metrics that describe valued ecological attributes (Ohio EPA 1989) and specific risk management strategies (Stevenson 1998). This invariably requires precise metric-stressor relationships to be defined. However, due to the relatively low number of non-wadeable rivers in Michigan, the complex nature of non-wadeable rivers, and the ways in which multiple stressors interact at different spatial scales (see Chapter 2), it is difficult to define single stressor-response relationships with the methods outlined in this chapter. This is the main weakness of my approach, and will require further examination.

The metrics selected for both NW-IBI assessment types have well-documented relationships to anthropogenic influences (Ohio EPA 1989; Karr and Chu 1999). While assessment of non-wadeable rivers will require more field as well as laboratory work than many of the currently used wadeable protocols, I believe that the NW-IBI, when used in conjunction with the Habitat Index (Wilhelm 2002; Wilhelm et al. 2005), provides an objective means of assessing the biological integrity of non-wadeable rivers in Michigan at the site scale.

The overall procedures outlined for assessing non-wadeable rivers in Michigan (Appendix 3) can be completed by a field team of 2 individuals in approximately 1 day, and depending on which type of assessment is chosen, macroinvertebrate processing may be conducted on site. The decision as to which type of assessment to use depends on the overall goal of the assessment and the number of transects with LWD.

The NW-IBI will undoubtedly require periodic fine-tuning and adjustment as additional data and experience arise. However, it appears to be a robust and sensitive means of evaluating the biotic integrity of Michigan's non-wadeable rivers. Future

research should be directed at refining metric selection criteria so as to enable the precise evaluation of stressors based on biological attributes, incorporating a fish procedure into the overall non-wadeable river assessment protocol. This will add a unique spatio-temporal dimension to the protocol and help MDEQ better-evaluate the ecological health of Michigan's non-wadeable river systems.

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TABLES

Table 3.1. List of sites sampled for macroinvertebrates.

Stream and site name	Code Name	Date Sampled	Stream and site name	Code Name	Date Sampled
Au Sable @ Whirlpool	as_whp01	08/14/01	Au Sable @ Mouth	as_mth02	07/30/02
Grand @ Comstock Riverside	gd_cmr01	07/18/01	Au Sable @ Whirlpool	as_whp02	07/31/02
Grand @ Grand Rapids	gd_gr01	07/19/01	Grand @ Comstock Riverside	gd_cmr02	06/13/02
Grand @ Ionia	gd_ion01	06/27/01	Grand @ Grand Haven	gd_gh02	06/19/02
Grand @ Johnson	gd_jon01	06/28/01	Grand @ Grand Ledge	gd_gld02	06/12/02
Kalamazoo @ Custer	kz_cus01	06/19/01	Grand @ Grand Rapids	gd_gr02	06/18/02
Kalamazoo @ Verberg	kz_ver01	06/21/01	Grand @ Ionia	gd_ion02	06/11/02
Manistee @ High Bridge	ma_hbr01	08/02/01	Grand @ Johnson	gd_jon02	06/18/02
Manistee @ Rainbow	ma_rbw01	08/01/01	Manistee @ Coates	ma_cts02	07/23/02
Muskegon @ Thornapple	mk_thp01	07/11/01	Manistee @ High Bridge	ma_hbr02	07/24/02
Muskegon @ Truckey	mk_trk01	07/10/01	Manistee @ Manistee	ma_mns02	07/25/02
Raisin @ Dundee	ra_dun01	07/04/01	Menominee @ Koss	me_kss02	07/09/02
Raisin @ Monroe	ra_mon01	07/03/01	Menominee @ Sturgeon Bend	me_stb02	07/10/02
Saginaw @ Zilwaukee	sg_zil01	07/31/01	Muskegon @ Big Rapids	mk_br02	06/25/02
Shiawassee @ Saginaw	sw_sg01	07/26/01	Muskegon @ Newaygo	mk_nwg02	06/26/02
Tittabawassee @ Saginaw	tb_sg01	08/07/01	St. Joseph @ Mottville	sj_mvl02	07/02/02
			St. Joseph @ Riverview	sj_rvw02	07/18/02
			Tahquamenon @ Newberry	tq_nwb02	07/12/02
			Tahquamenon @ Paradise	tq_pds02	07/13/02

Table 3.2. Catchment disturbance gradient (CDG) scores for each sample site. The seven individual measures were scored 0-4 with zero indicative of a natural system and 4 suggestive of a highly disturbed system. The scores for each metric were summed to give a total CDG score. See Table 3.1 for site codes. (Modified from Wilhelm 2002)

Site Code	% Urban, Buffer	% Ag, Buffer	% Urban, Basin	% Ag, Basin	Dam Density	NPDES Density	Road Density	*Total CDG Score*
tq_pds	0	0	0	0	0	0	0	*0*
ma_cts	0	0	0	0	1	0	0	*1*
ma_hbr	0	0	0	0	1	0	0	*1*
ma_rbw	1	0	0	0	1	0	0	*2*
me_stb	0	0	0	0	3	1	0	*3*
mk_thp	0	0	1	1	1	1	1	*3*
tq_nwb	0	0	0	0	3	0	1	*3*
as_whp	0	0	1	0	3	0	0	*4*
me_kss	2	0	0	0	3	1	0	*5*
mk_br	1	1	1	1	1	1	1	*5*
ma_mns	4	0	1	0	1	0	1	*6*
mk_nwg	1	2	1	1	1	1	2	*6*
as_mth	3	0	1	0	3	0	0	*7*
kz_cus	0	0	3	3	1	3	3	*7*
mk_trk	1	4	1	1	1	1	1	*8*
gd_gh	1	0	3	3	2	3	3	*9*
gd_jon	1	0	3	3	2	3	3	*9*
kz_alg	0	0	3	2	4	3	3	*9*
sw_sg	0	0	3	3	3	3	3	*9*
tb_sg	2	1	2	1	3	2	1	*9*
gd_ion	0	3	3	3	1	3	2	*10*
sg_zil	2	0	3	2	3	2	2	*10*
gd_gld	3	0	4	2	2	4	4	*11*
gd_gr	3	0	3	3	2	3	3	*11*
kz_con	4	0	3	3	1	3	3	*11*
ra_mon	3	0	2	4	2	3	3	*11*
sj_mvl	2	1	2	4	2	2	2	*11*
gd_cmr	4	0	3	3	2	3	3	*12*
gd_low	1	4	3	3	1	2	3	*12*
sj_rvw	3	1	2	4	2	2	3	*12*
kz_ver	4	0	4	2	3	3	4	*13*
ra_dun	3	2	2	4	2	3	3	*13*

Table 3.3. Total riparian disturbance gradient (RDG) score for each sample site based on the number of gaps in the riparian area and the mean riparian width. The two metrics were scored on a scale from 0-4 and were summed to yield a total score. A low number indicates a natural site, while a high number indicates a highly disturbed site. See Table 3.1 for site codes (Modified from Wilhelm 2002).

Site Code	Number of Gaps	Riparian Width	Total RDG Score
as_whp	0	0	0
ma_cts	0	0	0
me_stb	0	0	0
sw_sg	0	0	0
tq_nwb	0	0	0
tq_pds	0	0	0
ma_hbr	0	1	1
mk_nwg	0	1	1
gd_low	0	2	2
kz_cus	0	2	2
me_kss	1	1	2
gd_gh	1	2	3
gd_ion	1	2	3
kz_alg	2	1	3
mk_br	0	3	3
mk_trk	1	2	3
gd_jon	1	3	4
ma_rbw	2	2	4
mk_thp	2	2	4
sj_mvl	1	3	4
tb_sg	2	2	4
gd_gld	2	3	5
sg_zil	2	3	5
ra_dun	2	4	6
ra_mon	3	3	6
as_mth	4	3	7
gd_cmr	3	4	7
gd_gr	3	4	7
kz_ver	3	4	7
kz_con	4	4	8
ma_mns	4	4	8
sj_rvw	4	4	8

Table 3.4. List of macroinvertebrates found in Michigan non-wadeable rivers. Note that functional group assignments are at the family level and may vary within each family.

Order (or other higher taxon)	Family	Functional Feeding Group
Non-Insects		
Bivalvia	Sphaeriidae	CF
Bivalvia	Corbiculidae	CF
Bivalvia	Dreissenidae	CF
Bivalvia	Unionidae	CF
Gastropoda	Ancylidae	Sc
Gastropoda	Hydrobiidae	Sc
Gastropoda	Lymnaeidae	Sc
Gastropoda	Physidae	Sc
Gastropoda	Planorbidae	Sc
Gastropoda	Pleuroceridae	Sc
Gastropoda	Pomatiopsidae	Sc
Gastropoda	Valvatidae	Sc
Gastropoda	Viviparidae	Sc
Crustacea	Amphipoda	Sh
Crustacea	Argulidae	P
Crustacea	Decapoda	CG
Crustacea	Isopoda	Sh
Insecta		
Coleoptera	Carabidae	P
Coleoptera	Chrysomelidae	Sh
Coleoptera	Curculionidae	Sh
Coleoptera	Dytiscidae	P
Coleoptera	Elmidae	CG
Coleoptera	Gyrinidae	P
Coleoptera	Haliplidae	Pi
Coleoptera	Hydrophilidae	P
Coleoptera	Noteridae	P
Coleoptera	Psephenidae	Sc
Coleoptera	Scirtidae	Sc
Diptera	Athericidae	P
Diptera	Ceratopogonidae	P
Diptera	Chaoboridae	P
Diptera	Chironomidae	CG
Diptera	Culicidae	CF
Diptera	Dolichopodidae	P
Diptera	Empididae	P
Diptera	Ephydridae	Sh
Diptera	Muscidae	P

Table 3.4 (continued)

Order (or other higher taxon)	Family	Functional Feeding Group
Diptera	Simuliidae	CF
Diptera	Stratiomyidae	CG
Diptera	Tabanidae	P
Diptera	Tipulidae	CG
Ephemeroptera	Baetidae	CG
Ephemeroptera	Baetiscidae	CG
Ephemeroptera	Caenidae	CG
Ephemeroptera	Ephemerellidae	Sc
Ephemeroptera	Ephemeridae	CG
Ephemeroptera	Heptageniidae	Sc
Ephemeroptera	Isonychiidae	CF
Ephemeroptera	Leptophlebiidae	CG
Ephemeroptera	Polymitarcyidae	CG
Ephemeroptera	Potomanthidae	CF
Ephemeroptera	Trichorythidae	CG
Hemiptera	Belostomatidae	P
Hemiptera	Corixidae	
Hemiptera	Gerridae	P
Hemiptera	Hebridae	P
Hemiptera	Mesoveliidae	P
Hemiptera	Nepidae	P
Hemiptera	Notonectidae	P
Hemiptera	Pleidae	P
Hemiptera	Saldidae	P
Hemiptera	Veliidae	P
Hirudinea	Hirudinea	P
Hymenoptera	Mymaridae	P
Lepidoptera	Nepticulidae	Sh
Lepidoptera	Pyralidae	Sh
Megaloptera	Corydalidae	P
Megaloptera	Sialidae	P
Megaloptera	Sisyridae	P
Odonata	Aeshnidae	P
Odonata	Calopterygidae	P
Odonata	Coenagrionidae	P
Odonata	Corduliidae	P
Odonata	Gomphidae	P
Odonata	Lestidae	P

Table 3.4 (continued).

Order (or other higher taxon)	Family	Functional Feeding Group
Odonata	Libellulidae	P
Oligocheata	Oligochaeta	CG
Plecoptera	Perlidae	P
Plecoptera	Pteronarcyidae	Sh
Trichoptera	Brachycentridae	CF
Trichoptera	Dipseudopsidae	CF
Trichoptera	Glossosomatidae	Sc
Trichoptera	Helicopsychidae	Sc
Trichoptera	Hydropsychidae	CF
Trichoptera	Hydroptilidae	Sc
Trichoptera	Lepidostomatidae	Sh
Trichoptera	Leptoceridae	Sh
Trichoptera	Limnephilidae	Sh
Trichoptera	Philopotamidae	CF
Trichoptera	Phryganeidae	Sh
Trichoptera	Psychomyiidae	CG
Trichoptera	Uenoidae	Sc
Trichoptera	Polycentropodidae	P

Table 3.5. Potential biological attributes to be used as metrics in the final protocol. Potential metrics are categorized based on whether they are population, community, or functional attributes. Taxonomic resolution is to the family level. Asterisks indicate metrics retained for the composite assessment but not for the LWD assessment (EPT_RICH and P_RICH). Italics indicate metrics retained for the LWD assessment but not for the composite (SCR).

ATTRIBUTE	Code	Expected Response	Status	
			Corr	PCA
Population Level				
Ephemeroptera Richness	E_RICH	-	Discarded	Discarded
Plecoptera Richness	P_RICH	-	Retained	Retained*
Trichoptera Richness	T_RICH	-	Discarded	-
EPT Richness	EPT_RICH	-	Retained	Retained*
Diptera Richness	DIP_RICH	-	Retained	Retained
Community Level				
% Ephemeroptera	PER_E	-	Discarded	-
% Plecoptera	PER_P	-	Retained	Discarded
% Trichoptera	PER_T	-	Retained	Retained
% EPT	PER_EPT	-	Retained	Discarded
% Diptera	PER_DIP	+	Retained	Discarded
% Chironomidae	PER_CHIR	+	Discarded	-
% Oligochaeta	PER_OLIG	+	Retained	Discarded
Taxa Richness	RICH	-	Retained	Retained
Shannon Diversity	DIV	-	Discarded	-
% Dominance	PER_DOM	+	Retained	Retained
EPT/EPT+DIP	EPT_DIP	-	Discarded	-
Functional Group Metrics or Surrogates				
% Shredders	SHD	0	Retained	Discarded
% Scrapers	SCR	0	Retained	*Retained*
% Collector Filterers	CF	0	Retained	Discarded
% Collector Gatherers	CG	0	Retained	Discarded
% Predators	PRED	0	Retained	Discarded
FFG Diversity	FFG_DIV	-	Retained	Retained
Habitat Stability FFG*	HAB_STAB	-	Retained	Retained
P/R FFG	P_R	0	Discarded	-
CPOM:FPOM FFG	C_FPOM		Discarded	-
Transport:Benthic FPOM	T_BFPOM		Discarded	-

Table 3.6. Pearson correlation matrix of population level attributes with family-level data from composite samples. Bold numbers indicate highly correlated metrics (>0.70). Codes are listed in Table 3.5.

	E_RICH	P_RICH	T_RICH	EPT_RICH	DIP_RICH
E_RICH	1.000				
P_RICH	0.259	1.000			
T_RICH	0.413	0.280	1.000		
EPT_RICH	**0.826**	0.505	**0.821**	1.000	
DIP_RICH	0.255	0.121	**0.701**	0.540	1.000

Table 3.7. Pearson correlation matrix of population level attributes from LWD samples. Bold numbers indicate highly correlated metrics (>0.70). Taxonomic resolution is to the family level. Codes are listed in Table 3.5.

	E_RICH	P_RICH	T_RICH	EPT_RICH	DIP_RICH
E_RICH	1.000				
P_RICH	0.383	1.000			
T_RICH	0.488	0.371	1.000		
EPT_RICH	**0.812**	0.585	**0.883**	1.000	
DIP_RICH	0.386	-0.027	0.523	0.478	1.000

Table 3.8. Pearson correlation matrix of community level attributes with family-level data from composite samples. Bold numbers indicate highly correlated metrics (>0.70). Codes are listed in Table 3.5.

	RICH	PER_DOM	EPT_DIP	DIV	PER_DIP	PER_CHIR	PER_OLG	PER_E	PER_P	PER_T	PER_EPT
RICH	1.000										
PER_DOM	-0.596	1.000									
EPT_DIP	0.389	-0.474	1.000								
DIV	**0.750**	**-0.940**	0.496	1.000							
PER_DIP	-0.210	0.396	-0.348	-0.387	1.000						
PER_CHIR	-0.425	**0.785**	-0.698	**-0.703**	0.503	1.000					
PER_OLIG	-0.357	0.215	-0.563	-0.320	0.065	0.298	1.000				
PER_E	0.119	-0.163	**0.704**	0.168	-0.103	-0.312	-0.306	1.000			
PER_P	-0.018	-0.234	0.459	0.240	-0.081	-0.288	-0.230	0.205	1.000		
PER_T	0.106	-0.029	0.487	0.092	-0.236	-0.185	-0.326	-0.070	0.391	1.000	
PER_EPT	0.162	-0.158	**0.888**	0.201	-0.234	-0.378	-0.459	**0.769**	0.446	0.583	1.000

Table 3.9. Pearson correlation matrix of community level attributes from LWD samples. Bold numbers indicate highly correlated metrics (>0.70). Taxonomic resolution is to the family level. Codes are listed in Table 3.5.

	RICH	PER_DOM	EPT_DIP	DIV	PER_DIP	PER_CHIR	PER_E	PER_P	PER_T	PER_EPT
RICH	1.000									
PER_DOM	-0.575	1.000								
EPT_DIP	0.536	**-0.782**	1.000							
DIV	0.471	**-0.913**	**0.774**	1.000						
PER_DIP	-0.495	**0.872**	**-0.897**	**-0.849**	1.000					
PER_CHIR	-0.340	0.267	-0.282	-0.260	0.277	1.000				
PER_E	0.313	-0.545	0.666	0.595	-0.575	-0.138	1.000			
PER_P	0.244	-0.336	0.411	0.422	-0.434	-0.185	0.268	1.000		
PER_T	0.426	-0.355	0.649	0.304	-0.479	-0.185	-0.059	0.136	1.000	
PER_EPT	0.546	-0.654	**0.961**	0.652	**-0.773**	-0.243	0.642	0.336	**0.726**	1.000

Table 3.10. Pearson correlation matrix for functional attributes with family-level data from composite samples. Bold numbers indicate highly correlated metrics (>0.70). Codes are listed in Table 3.5.

	SHD	SCR	CF	CG	PRED	P_R	HAB_ STAB	C_FPOM	T_B FPOM	FFG_ DIV
SHD	1.000									
SCR	0.053	1.000								
CF	0.043	0.090	1.000							
CG	-0.465	-0.638	-0.556	1.000						
PRED	-0.213	-0.148	-0.039	0.117	1.000					
P_R	-0.034	**0.963**	0.065	-0.593	-0.119	1.000				
HAB_STAB	-0.052	0.684	0.674	**-0.728**	-0.058	0.688	1.000			
C_FPOM	**0.907**	0.093	-0.049	-0.429	-0.153	0.026	-0.036	1.000		
T_BFPOM	0.026	0.227	**0.915**	-0.609	-0.051	0.205	**0.836**	-0.017	1.000	
FFG_DIV	0.457	0.587	0.470	**-0.805**	0.026	0.455	0.513	0.310	0.403	1.000

Table 3.11. Pearson correlation matrix for functional attributes from LWD samples. Bold numbers indicate highly correlated metrics (>0.70). Taxonomic resolution is to the family level. Codes are listed in Table 3.5.

	SHD	SCR	CF	CG	PRED	P_R	HAB_STAB	C_FPOM	T_BFPOM	FFG_DIV
SHD	1.000									
SCR	-0.036	1.000								
CF	0.224	-0.069	1.000							
CG	-0.346	-0.282	-0.509	1.000						
PRED	0.038	0.251	-0.184	-0.357	1.000					
P_R	0.005	**0.912**	-0.145	-0.472	0.466	1.000				
HAB_STAB	0.057	0.225	**0.836**	-0.664	0.039	0.220	1.000			
C_FPOM	0.446	-0.009	-0.024	-0.579	0.373	0.344	0.081	1.000		
T_BFPOM	0.201	-0.106	0.933	-0.577	-0.093	-0.116	**0.916**	0.066	1.000	
FFG_DIV	0.453	0.619	0.457	-0.650	0.346	0.570	0.463	0.211	0.331	1.000

Table 3.12. Results of principal component analysis for both composite and large woody debris (LWD) samples. Bold values indicate axes that were further examined for metric selection.

PCA Axis	Eigenvalues	
	Composite	*LWD*
1	**6.51766**	**6.28627**
2	**2.32852**	**1.86367**
3	**1.92508**	**1.52759**
4	**1.34371**	**1.0996**
5	**1.04998**	0.906779
6	0.838963	0.670256
7	0.803271	0.342908
8	0.567636	0.269082
9	0.398593	0.249948
10	0.218721	0.163461
11	0.212564	0.07233
12	0.140631	0.054165
13	0.07073	0.011848
14	0.059754	0.005174
15	0.033363	0.005089
16	0.01015	0
17	0.003563	0

Table 3.13. Principal component loadings for composite samples. In general, those metrics with the highest loadings per axis were retained for the final protocol. When two or more metrics had similarly high loadings, two were chosen for the final protocol. Bold numbers indicate which metrics were retained from each axis. Some metrics with high loadings were not retained due to inaccuracy (e.g., PER_OLIG). See Table 3.5 for metric codes.

	Axis 1	Axis 2	Axis 3	Axis 4	Axis 5
PRICH	0.428	0.486	-0.172	**-0.467**	-0.062
EPTRICH	0.586	0.108	**-0.652**	-0.070	-0.290
DIPRICH	0.500	-0.180	-0.569	**0.410**	0.083
RICH	0.682	-0.239	**-0.625**	0.084	-0.076
PERDMC	-0.785	0.331	0.048	0.200	**-0.345**
PERDIP	-0.764	0.170	-0.257	0.112	-0.253
PEROLIG	-0.612	-0.105	-0.122	-0.149	0.619
PERP	0.430	0.588	0.239	-0.529	0.035
PERT	0.426	**0.713**	0.077	0.457	0.119
PEREPT	0.452	0.578	0.052	0.051	-0.266
LNSHD	0.560	-0.312	-0.025	0.085	-0.020
LNSCR	0.580	-0.493	0.285	-0.213	-0.377
CF	0.637	0.456	0.135	0.440	0.265
CG	-0.853	0.184	-0.312	-0.046	-0.195
LNPRED	0.138	0.284	-0.578	-0.391	0.258
LNHSTAB	**0.839**	-0.011	0.362	0.052	-0.059
FFGDIV	**0.905**	-0.235	0.052	-0.103	0.093

Table 3.14. Principal component loadings for LWD samples. In general, those metrics with the highest loadings per axis were retained for the final protocol. When two or more metrics had similarly high loadings, 2-3 were chosen for the final protocol. Bold numbers indicate which metrics were retained from each axis. Some metrics with high loadings were not retained due to ambiguous response to human influences (e.g., LNSH) See Table 3.5 for metric codes.

	Axis 1	Axis 2	Axis 3	Axis 4
RICH	0.7826	-0.2029	-0.4415	0.1764
PRICH	0.5499	0.1234	-0.00821	-0.2297
EPTRICH	0.7727	-0.3737	-0.2902	0.01024
PERCG	-0.6977	-0.4215	-0.3727	0.01761
DIPRICH	0.3951	-0.3218	**-0.5372**	0.5403
PERDOM	**-0.813**	-0.2592	0.0859	0.05418
FFGDIV	**0.8592**	0.3726	-0.04755	0.1654
PEREPT	0.7988	-0.2562	0.1289	-0.2365
LNSC	0.6534	0.04705	-0.4381	**-0.4052**
LNCF	0.6487	-0.3167	0.5203	0.3262
LNHABSTAB	**0.8653**	0.07889	0.3857	-0.1131
LNPRED	0.2839	0.6564	-0.3485	-0.2277
LNSH	0.1998	0.6841	0.08521	0.5824
LNTRICH	0.7677	**-0.4241**	0.3334	-0.04375

Table 3.15. Metrics retained and their corresponding weights in the final protocol (composite samples only). Weights were determined by summing eigenvalues from retained axes and calculating the total variance explained by each axis. Scores were then approximated based on a 100-point scale. When 2 or more metrics were retained from each axis, scores were divided equally based on the variance calculated for the entire axis. See Table 3.5 for metric codes.

Metric	Axis	Eigenvalue	Variance (Prop/Total)	Score
FFG_DIV	1	6.51766	0.50	25
HAB_STAB	1			25
PER_T	2	2.32852	0.18	20
EPT_RICH	3	1.92508	0.15	8
RICH	3			7
DIP_RICH	4	1.34371	0.10	5
P_RICH	4			5
PER_DOM	5	1.04998	0.08	5
	Total	13.16495		100

Table 3.16. Metrics retained and their corresponding weights in the final protocol (LWD samples only). Weights were determined by summing eigenvalues from retained axes and calculating the total variance explained by each axis. Scores were then approximated based on a 100 point scale. When 2 or more metrics were retained from each axis, scores were divided equally based on the variance calculated for the entire axis. See Table 3.5 for metric codes.

Metric	Axis	Eigenvalue	Variance (Prop/Total)	Score
HAB_STAB	1			20
FFG_DIV	1	6.28627	0.58	20
PER_DOM	1			20
PER_T	2	1.86367	0.17	15
DIP_RICH	3	1.52759	0.14	15
SCR	4	1.09960	0.10	10
	Total	10.77713		100

Table 3.18. Scoring criteria for biological monitoring of non-wadeable rivers using large woody-debris samples only.

Biological Metric	Poor	Fair	Good	Excellent	Metric Total
Habitat Stability FFG Surrogate	Hab Stab <0.08	Hab Stab 0.08-0.19	Hab Stab 0.20-0.56	Hab Stab >0.56	
	0	7	14	20	
FFG Diversity	FFG Diversity <0.76	FFG Diversity 0.76-1.09	FFG Diversity 1.10-1.33	FFG Diversity >1.33	
	0	7	14	20	
% Dominant Taxon	% Dominance >59	% Dominance 34-59	% Dominance 23-33	% Dominance <23	
	0	7	14	20	
% Trichoptera Abundance	% Trichoptera <3	% Trichoptera 3-6	% Trichoptera 7-14	% Trichoptera >14	
	0	5	10	15	
Diptera Taxa Richness	Diptera Richness <2	Diptera Richness 2-3	Diptera Richness 4-5	Diptera Richness >5	
	0	5	10	15	
% Scraper Abundance	% Scrapers <2	% Scrapers 2-6	% Scrapers 7-11	% Scrapers >11	
	0	4	7	10	
Total Point Score					

Table 3.17. Scoring criteria for biological monitoring of non-wadeable rivers using composite samples with all habitats.

Biological Metric	Poor	Fair	Good	Excellent	Metric Total
FFG Diversity	FFG Div <0.95	FFG Div 0.95-1.41	FFG Div 1.42-1.70	FFG Div >1.71	
	0	8	16	25	
Habitat Stability FFG Surrogate	Hab Stab <0.09	Hab Stab 0.09-0.26	Hab Stab 0.27-0.67	Hab Stab >0.67	
	0	8	16	25	
% Trichoptera	% Trichoptera <1.3	% Trichoptera 1.30-3.40	% Trichoptera 3.41-6.80	% Trichoptera >6.80	
	0	7	14	20	
EPT Richness	EPT Rich <4	EPT Rich 4-6	EPT Rich 7-9	EPT Rich >9	
	0	3	6	8	
Total Richness	Taxa Richness <15	Taxa Richness 15-18	Taxa Richness 19-24	Taxa Richness >24	
	0	2	5	7	
Diptera Richness	Dip Richness	Dip Richness	Dip Richness	Dip Richness	
	<2	2-3	4-5	>5	
	0	2	4	5	
Plecoptera Richness	Plec. Richness	Plec Richness	Plec Richness	Plec Richness	
	0	1	2	3	
	0	2	4	5	
% Dominance	% Dominance	% Dominance	% Dominance	% Dominance	
	>60	47-60	35-46	<35	
	0	2	4	5	
Total Point Score					

Table 3.19. Results from multiple linear regression of composite metrics with environmental variables. Forward stepwise regression (p-value to enter=0.15; tolerance=0.001) and backward stepwise regression (p-value to remove=0.15; tolerance 0.001) sometimes selected different environmental variables to which metrics respond. Only the first 4 variables to enter (forward) or be removed (backward) are shown here. [1]General response to riparian landuse. [2]General response to watershed landuse. *Denotes variables with $p<0.05$. **Denotes variables with $p<0.001$. In all cases, $p<0.10$. See Table 3.5 for metric codes and Tables 19-20 for environmental variable codes.

Metric	Method	X_1	X_2	X_3	X_4	R^2
FFG_DIV[1]	Forward	DO	TURB*	CHL	NatRP*	0.62
	Backward	TP*	COND*	TURB*	CHL*	0.75
HAB_STAB	Forward	COND*	AgRP*	--	--	0.37
	Backward	UrWS*	AgWS*	NatWS*	AgRP*	0.56
PER_T	Forward	TURB	AgRP*	--	--	0.64
	Backward	TOT N	AgRP*	--	--	0.64
EPT_RICH	Forward	LWD*	UrWS*	--	--	0.31
	Backward	LWD*	UrWS*	--	--	0.31
RICH	Forward	UrWS*	--	--	--	0.19
	Backward	UrWS	NatWS		--	0.22
DIP_RICH	Forward	TOT N*	PH	TURB	--	0.33
	Backward	TOT N*	PH*	TURB*	CHL*	0.66
P_RICH	Forward	LWD**	--	--	--	0.39
	Backward	LWD**	PH	AgWS	NatWS	0.49
PER_DOM[1,2]	Forward	DO	CHL	NatRP*	--	0.46
	Backward	TP*	COND*	DO	TURB	0.69

Table 3.20. Results from multiple linear regression of LWD metrics with environmental variables. Forward stepwise regression (p-value to enter=0.15; tolerance=0.001) and backward stepwise regression (p-value to remove=0.15; tolerance 0.001) sometimes selected different environmental variables to which metrics respond. Only the first 4 variables to enter (forward) or be removed (backward) are shown here. [1]General response to riparian landuse. *Denotes variables with $p<0.05$. **Denotes variables with $p<0.001$. In all cases, $p<0.10$. See Table 3.5 for metric codes and Tables 19-20 for environmental variable codes.

Metric	Method	X_1	X_2	X_3	X_4	R^2
HAB_STAB	Forward	NatWS**	AgRP	--	--	0.48
	Backward	TOT N*	AgRP*	NatRP*	--	0.53
FFG_DIV	Forward	PH*	UrWS*	NatRP	--	0.66
	Backward	TOT N**	TP*	PH**	NatRP*	0.75
PER_DOM	Forward	DO*	UrWS*	NatRP**	--	0.75
	Backward	DO*	UrWS*	NatRP**	--	0.75
PER_T	Forward	PH*	AgWS*	AgRP*	--	0.53
	Backward	PH*	UrWS*	AgRP*	--	0.55
DIP_RICH	Forward	TOT N*	UrWS**	NatWS**	UrRP*	0.71
	Backward	TOT N*	UrWS**	NatWS**	UrRP*	0.71
SCR[1]	Forward	NatWS**	--	--	--	0.46
	Backward	TP*	COND*	PH*	TURB	0.76

Table 3.21. Results from the discriminant function analysis. Two analyses were done: One in which approximately one-half of the sites from each classification type were used to generate the model, while the other half were used to test the model. The other analysis used jackknifing to evaluate the model.

Classificaiton	**% Correct (test sites only)**		**% Correct (jackknifed)**	
	Composite	*LWD*	*Composite*	*LWD*
Poor	75	100	88	78
Fair	33	25	50	75
Good	67	100	55	60
Excellent	100	33	67	50

Table 3.22. Site rankings based on CDG, RDG, and an overall ranking based on CDG and RDG rankings combined with the number of transects with LWD (Mean Rank). HI=Non-wadeable Habitat Index (Wilhelm 2002). Site scores for composite and LWD assessments are also listed. These data were used to evaluate the NW-IBI sensitivity (composite vs. LWD) to differing scales of human impacts.

Site	CDG Rank	RDG Rank	Mean Rank	HI	NW IBI Comp	NW-IBI LWD
as_mth02	30	20	17	47	69	47
as_whp01	1	13	1	83	69	57
as_whp02	3	21	4	86	34	77
gd_cmr01	28	28	29	25	27	16
gd_cmr02	31	27	32	26	29	4
gd_gh02	16	17	19	43	2	5
gd_gld02	25	18	27	50	48	37
gd_gr01	29	31	28	27	27	27
gd_gr02	32	30	33	32	29	35
gd_ion01	13	11	22	51	42	21
gd_ion02	17	4	24	59	36	5
gd_jon01	14	1	13	52	63	54
gd_jon02	22	5	18	51	31	19
kz_cus01	11	2	11	66	62	59
ma_cts02	4	22	5	85	84	80
ma_hbr01	8	14	3	82	70	69
ma_hbr02	9	6	6	85	69	89
ma_mns02	33	25	21	28	58	19
ma_rbw01	19	12	10	66	74	95
me_kss02	12	19	12	58	45	71
me_stb02	5	23	7	69	77	89
mk_br02	18	7	15	49	9	32
mk_nwg02	10	8	14	60	56	61
mk_thp01	20	15	8	78	65	65
mk_trk01	15	16	20	50	67	63
ra_dun01	26	3	30	38	17	31
ra_mon01	27	32	31	34	8	0
sg_zil01	24	29	25	31	0	5
sh__sg01	2	33	16	63	22	.N/A
sj_mvl02	23	9	26	66	79	64
sj_rvw02	34	24	34	39	20	38
tb_sag01	21	26	23	42	22	40
tq_nwb02	6	34	9	59	40	.N/A
tq_pds02	7	10	2	59	40	63

Table 3.23. Regression data for composite assessments. In all cases, the composite NW-IBI was the dependent variable. . Independent variables reflect differing scales of human influence. CDG=Catchment Disturbance Gradient; RDG=Riparian Disturbance Gradient; Mean Rank is based on catchment-wide, riparian, and in-stream habitat quality. HI=Non-wadeable Habitat Index (Wilhelm 2002)

Independent Variable	R^2	P-value
CDG	0.130	0.036
RDG	0.113	0.052
Mean Rank	0.313	<0.001
HI	0.407	<0.001

Table 3.24. Regression data for LWD assessments. In all cases, the LWD NW-IBI was the dependent variable. Independent variables reflect differing scales of human influence. CDG=Catchment Disturbance Gradient; RDG=Riparian Disturbance Gradient; Mean Rank is based on catchment-wide, riparian, and in-stream habitat quality. HI=Non-wadeable Habitat Index (Wilhelm 2002)

Independent Variable	R^2	P-value
CDG	0.338	<0.001
RDG	0.083	0.110
Mean Rank	0.535	<0.001
HI	0.612	<0.001

FIGURES

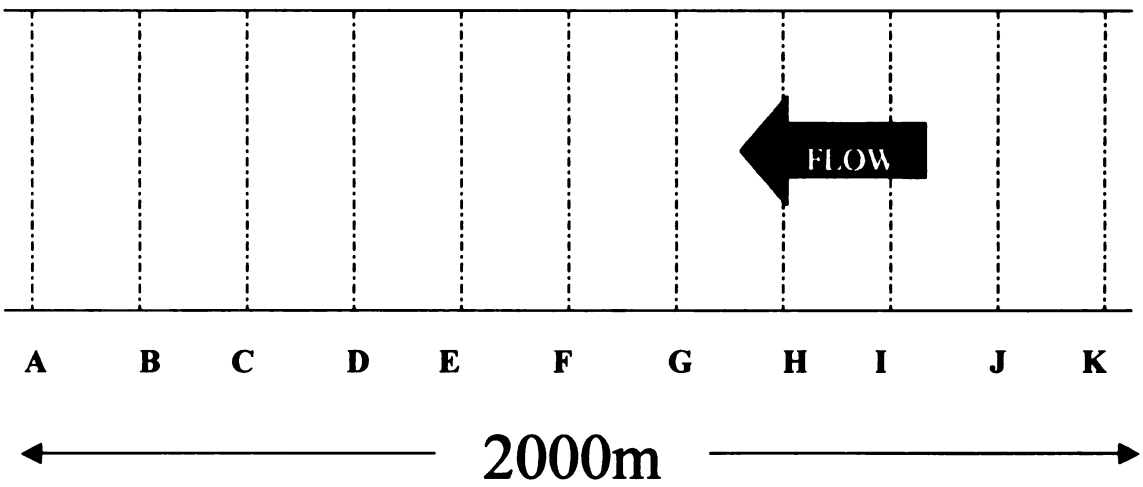


Figure 3.1. Diagram of a non-wadeable study reach. I chose a standard 2000m reach, and sampled macroinvertebrates at transects placed every 200m.

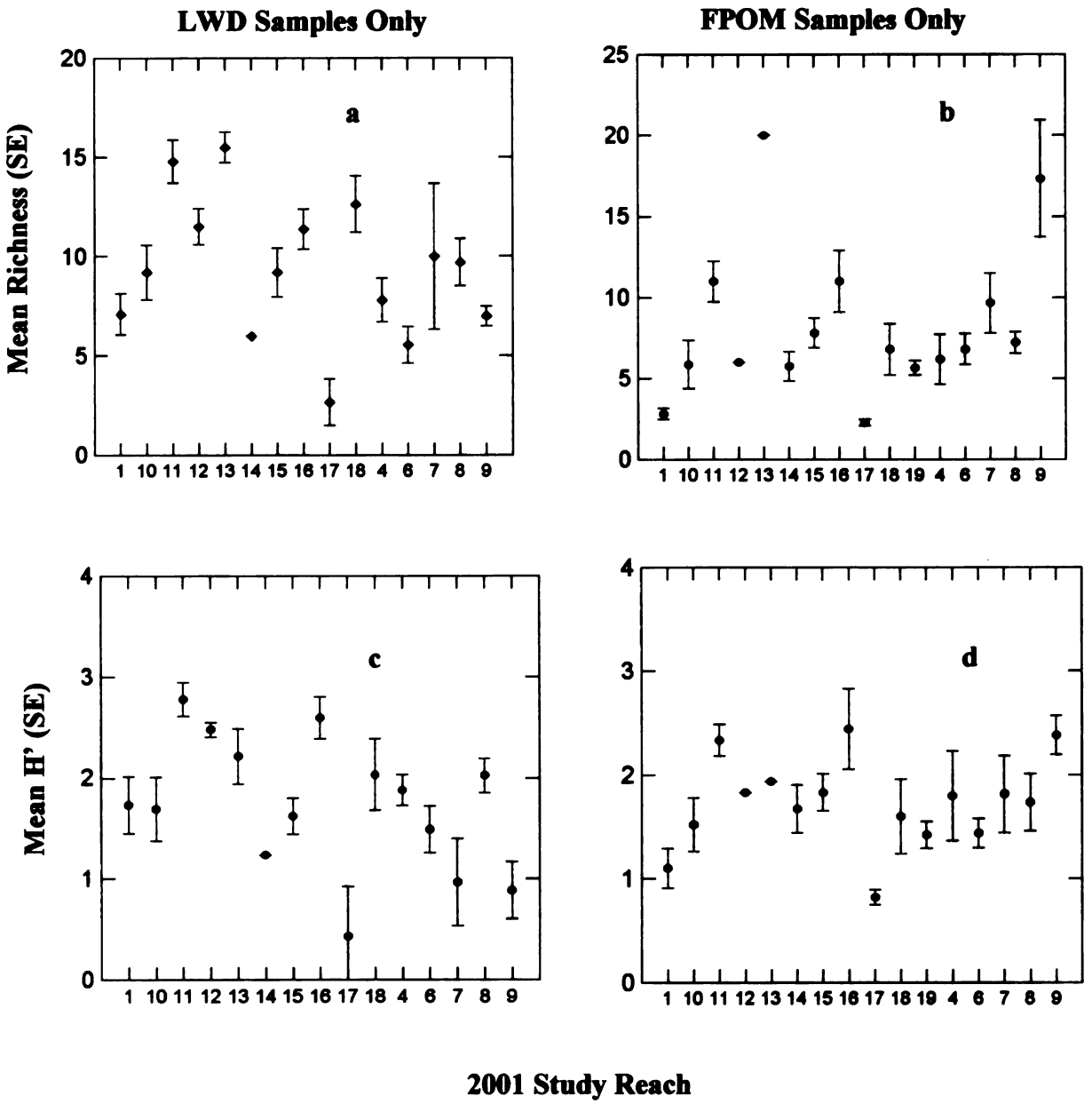


Figure 3.2. Mean taxa richness and Shannon diversity (H') from selected 2001 study sites. LWD (a and c) and FPOM (b and d) samples only. Note that despite coming from the same habitat, richness and diversity still varied considerably in LWD samples, while variability was less pronounced in FPOM samples. Numbers on x-axis indicate different sites.

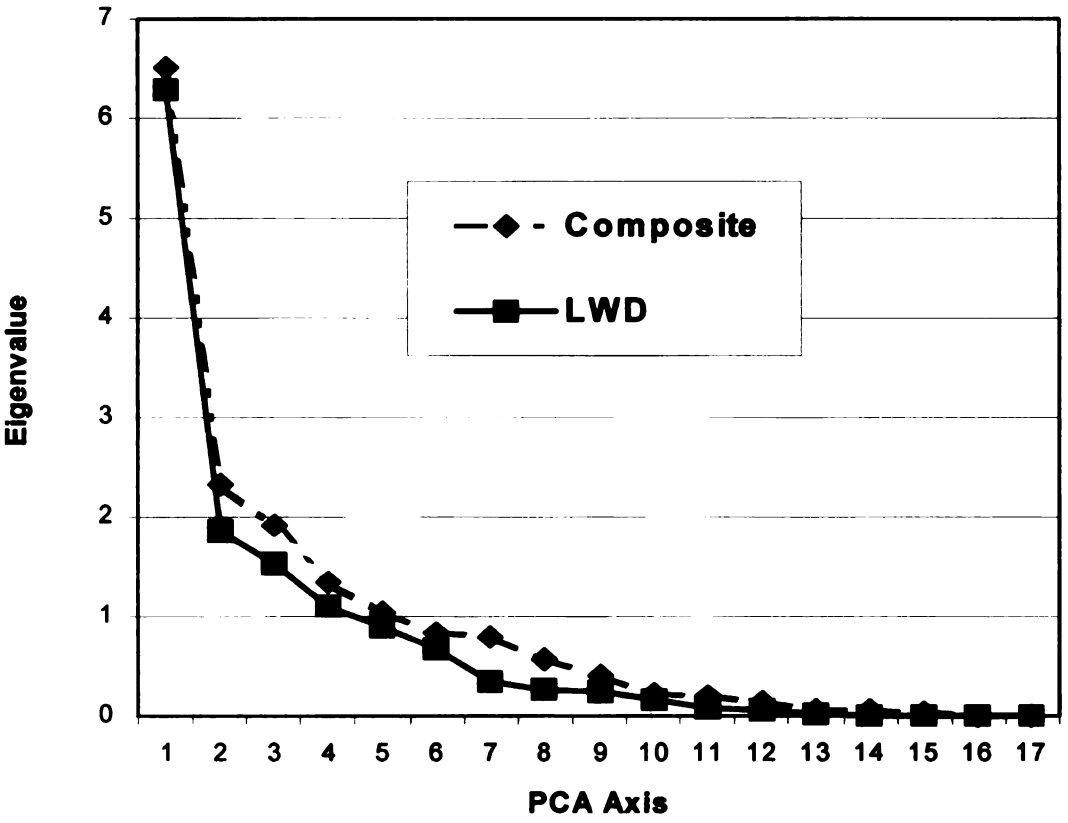


Figure 3.3. Scree plot of eigenvalues (λ) for each PCA axis. Axes 1-5 were further examined for composite metric selection. Axes 1-4 were further examined for LWD metric selection. Criteria: λ>1.

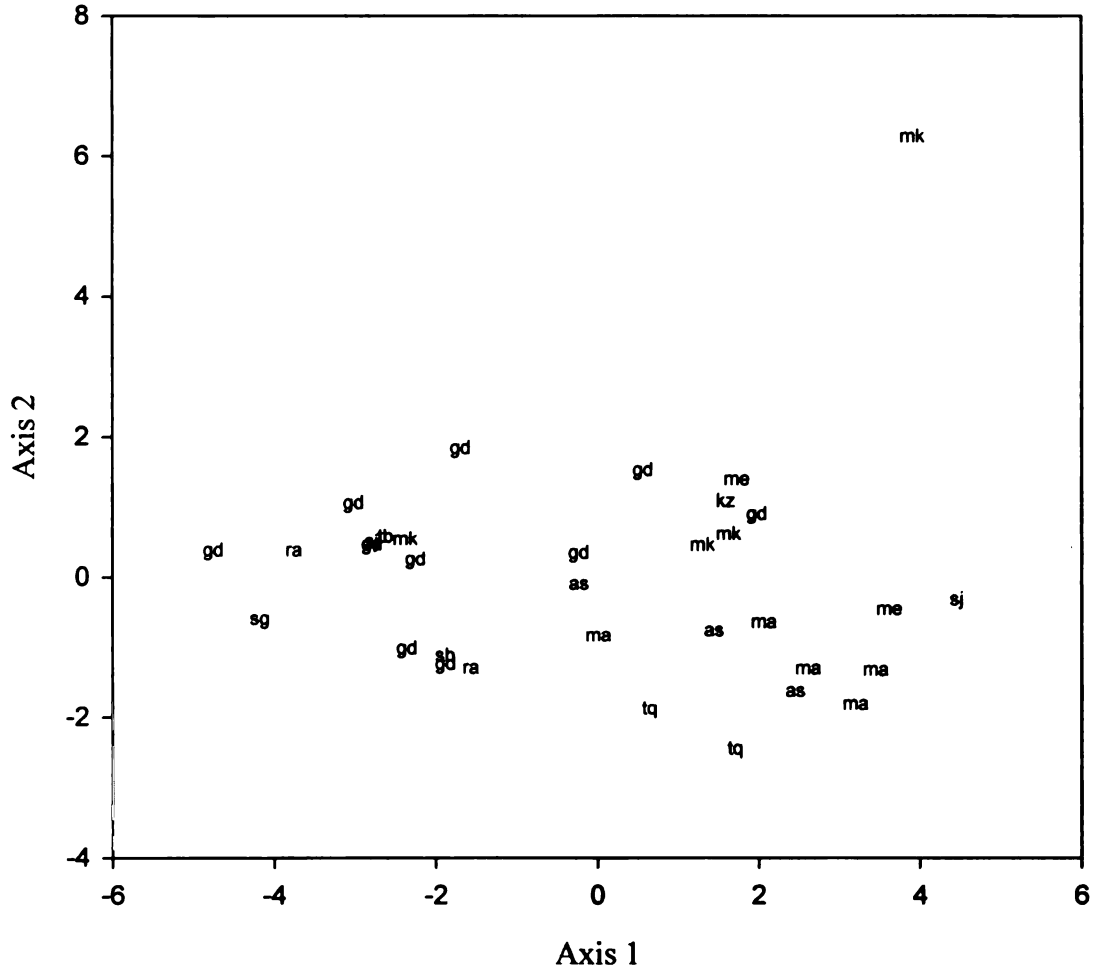


Figure 3.4. PCA site scores (axes 1 vs. 2) for the composite data. Site scores are plotted with rivers identified (see Table 3.1 for river codes). This plot shows that sites did not cluster by river or catchment.

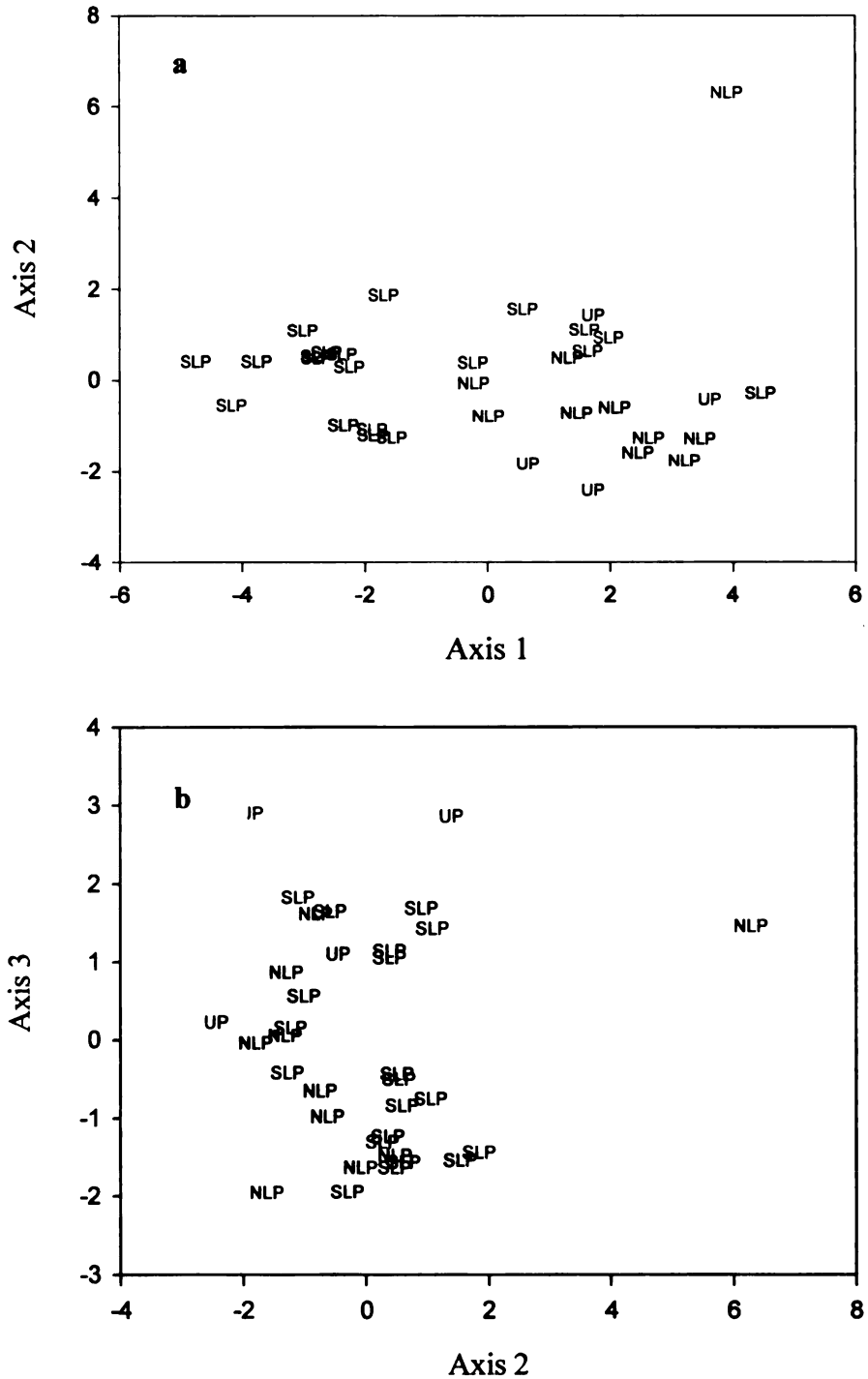


Figure 3.5. PCA site scores identified by ecoregion. (a) Axis 1 vs. Axis 2; (b) Axis 2 vs. Axis 3. Sites clustered based on ecoregion along axis 1, but not along the other axes. SLP: Southern Lower Peninsula; NLP: Northern Lower Peninsula; UP: Upper Peninsula.

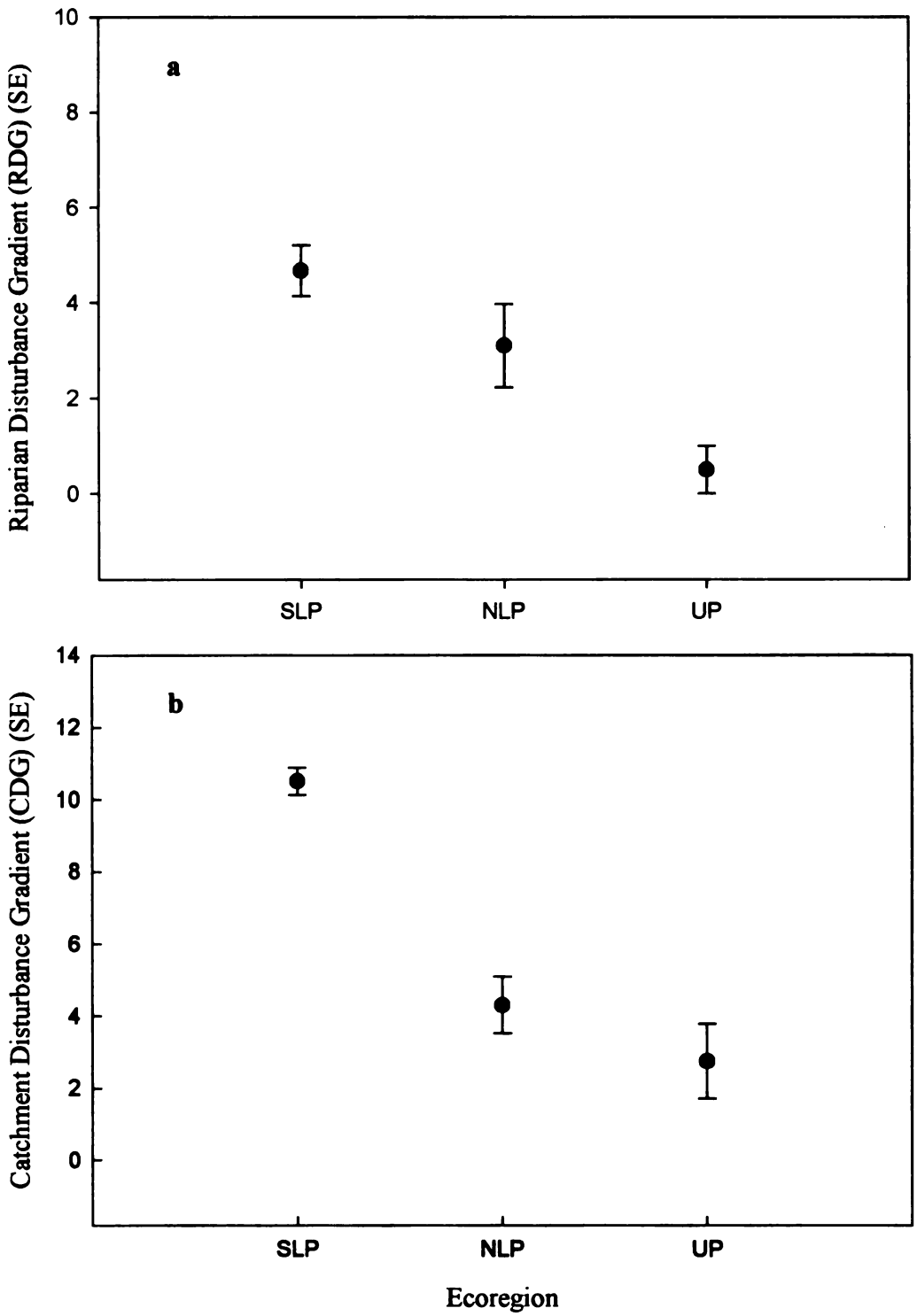


Figure 3.6. (a) Mean RDG and (b) mean CDG by ecoregion (± SE) (See Wilhelm 2002 for information regarding the development of the RDG and CDG).

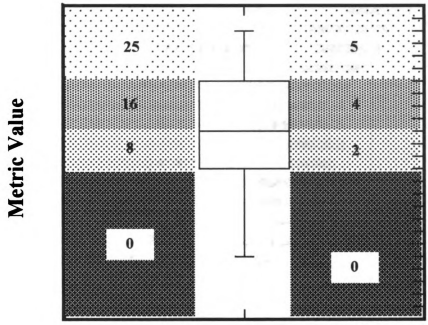


Figure 3.7. Theoretical example of how a 25 point and a 5 point metric were scored based on inter-quartile ranges.

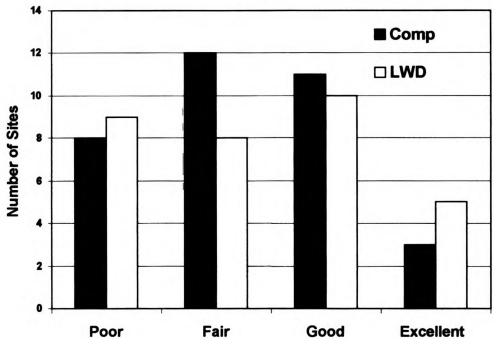


Figure 3.8. NW-IBI scores for all sites comparing composite and LWD assessments. LWD assessments appeared to be more sensitive than composite assessments.

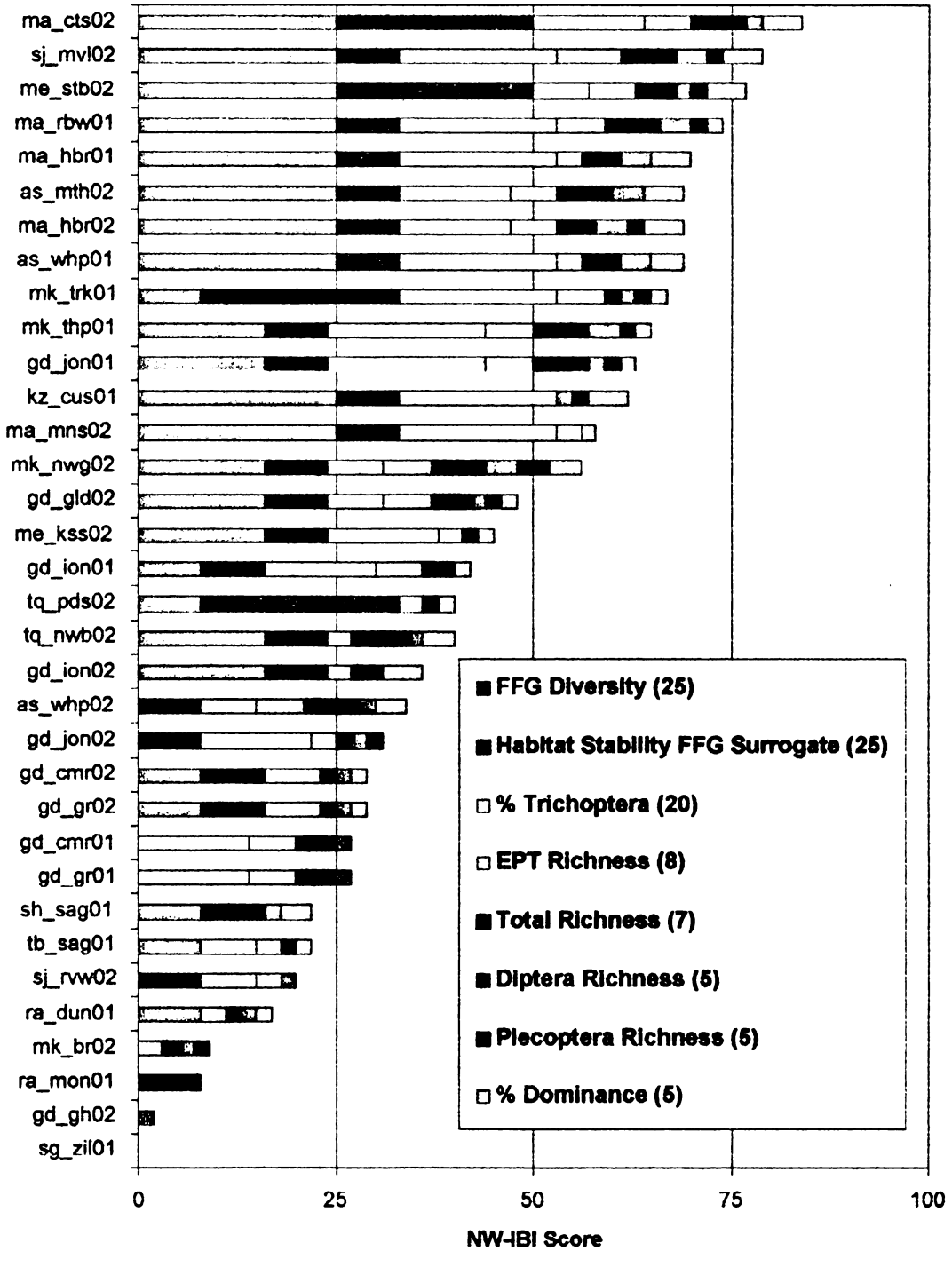


Figure 3.9. Composite NW-IBI scores for each non-wadeable study site. Individual metric scores are shown in each bar. This image is presented in color. See Table 3.1 for site codes.

Figure 3.9: Composite IVW-TBI scores for each non-wadeable study site. Individual metric scores are shown in each bar. This image is presented in color. See Table 3.1 for site codes.

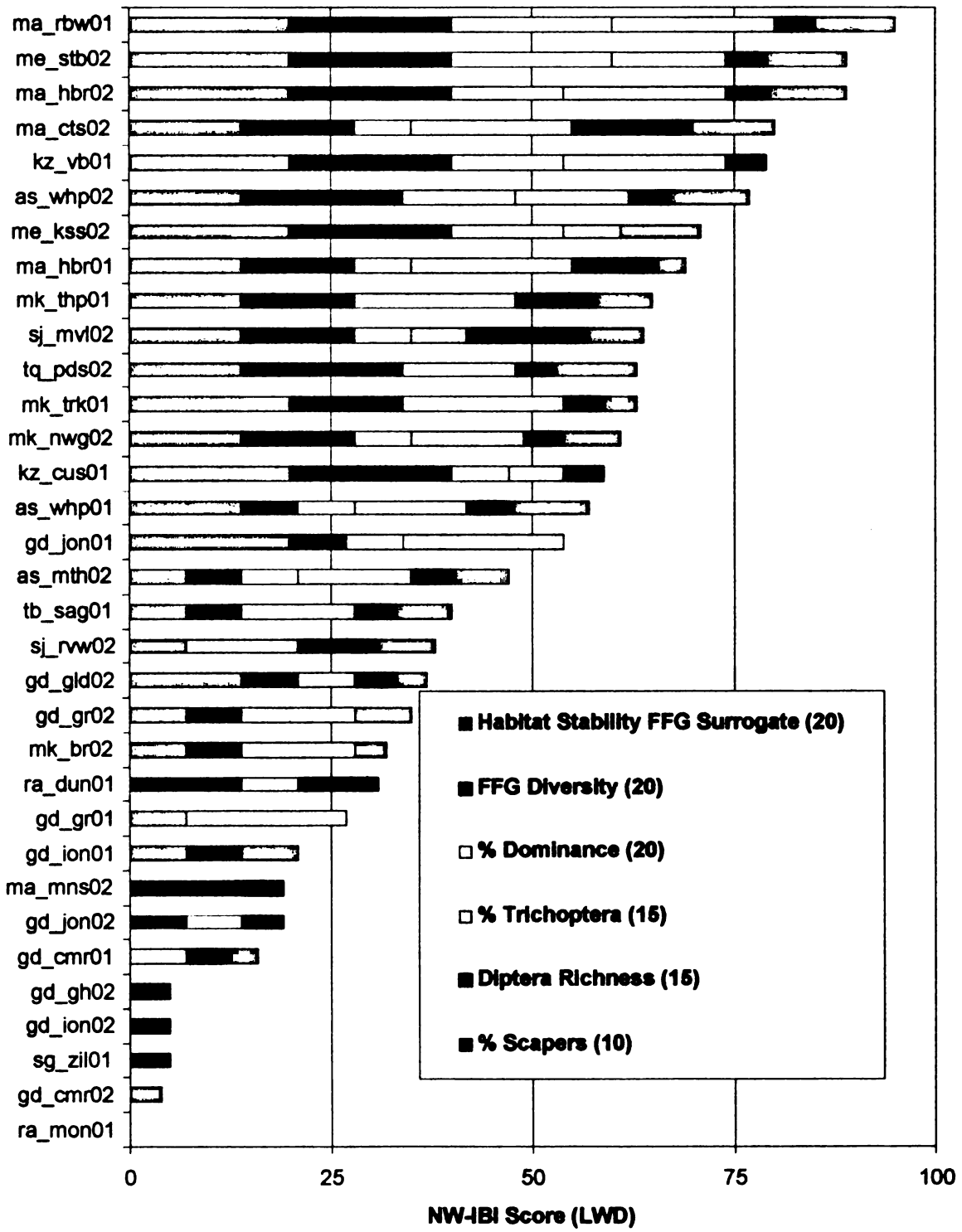


Figure 3.10. LWD NW-IBI scores for each non-wadeable study site. Individual metric scores are shown in each bar. This image is presented in color. See Table 3.1 for site codes.

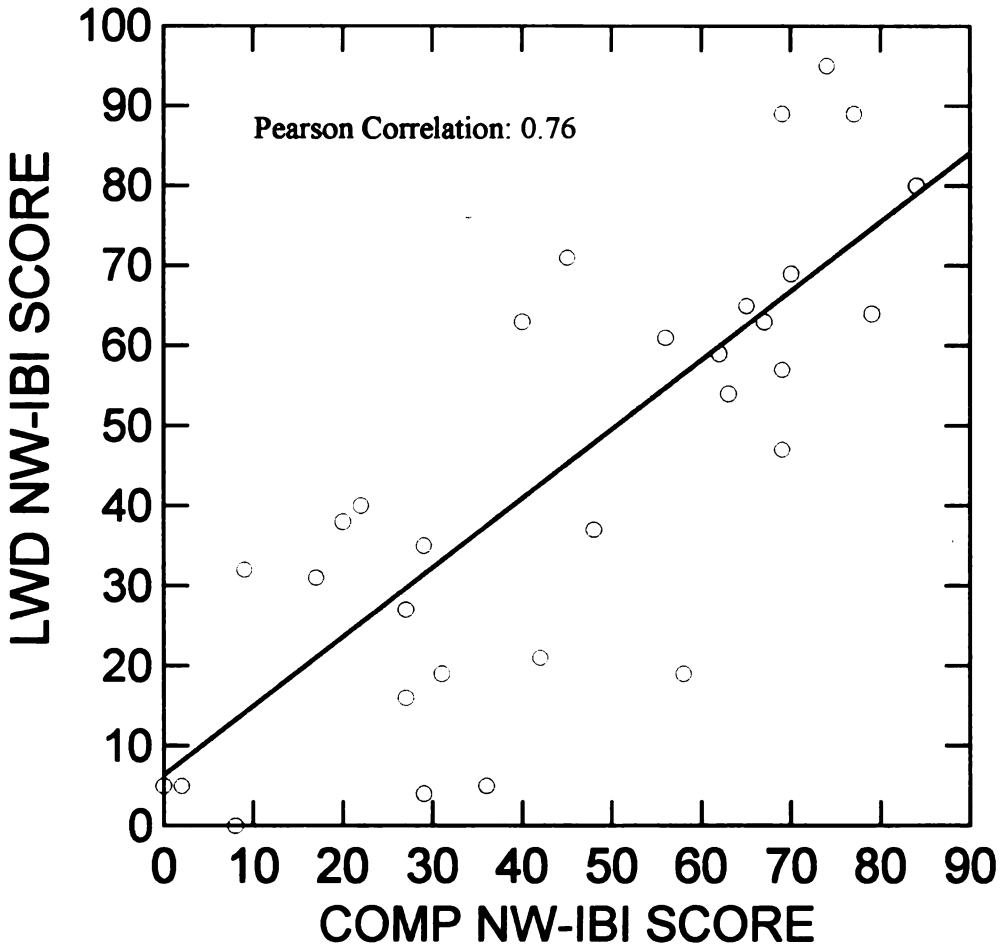


Figure 3.11. Composite vs. LWD NW-IBI scores for non-wadeable river study sites.

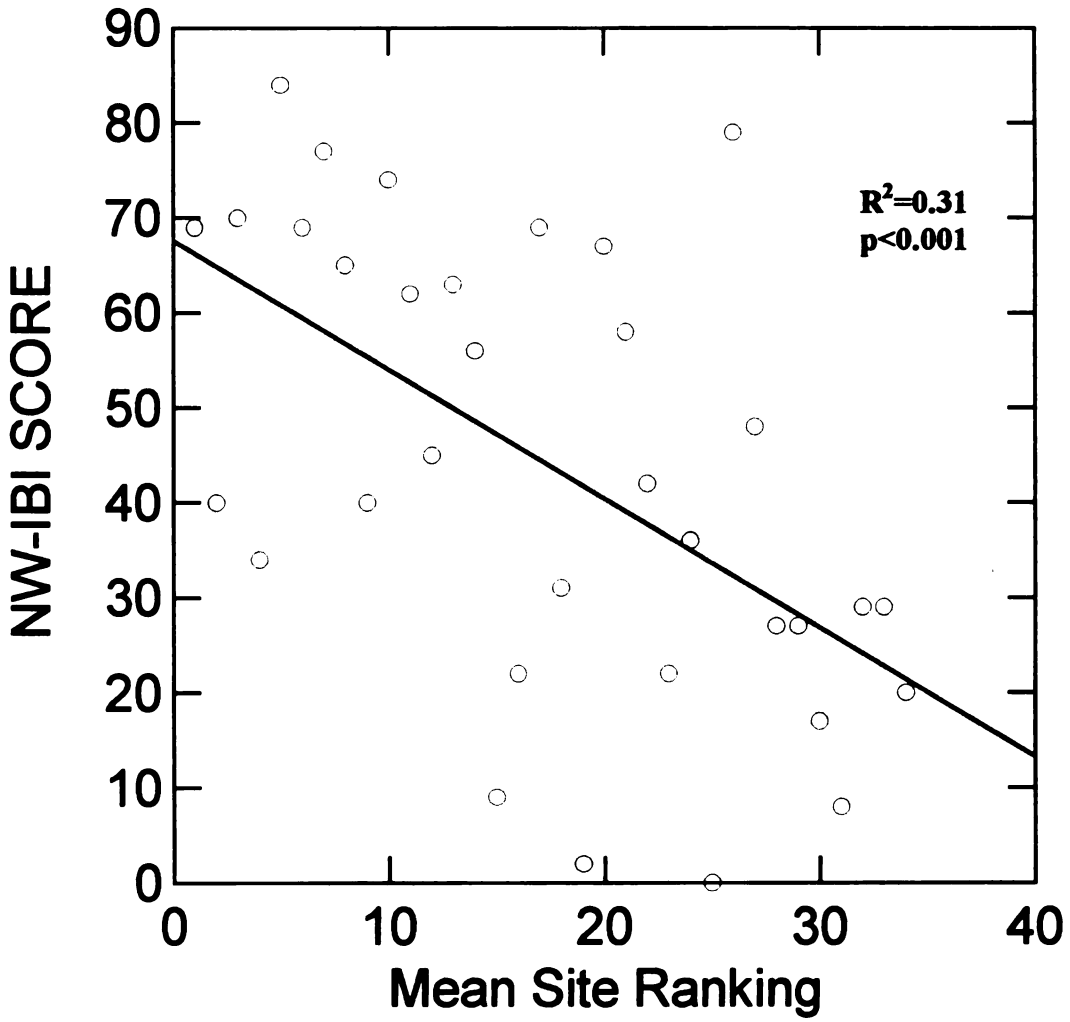


Figure 3.12. Mean Site Ranking vs. NW-IBI for composite assessments.

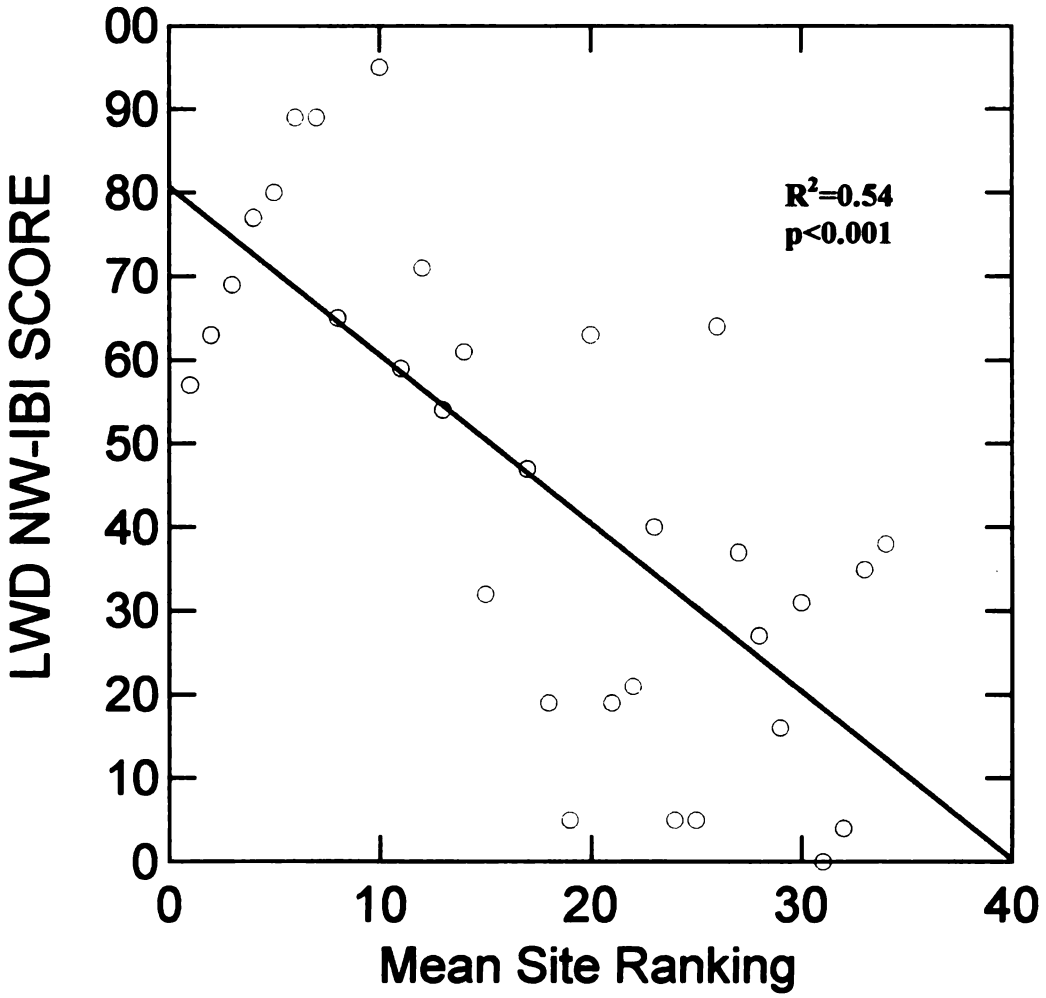


Figure 3.13. Mean Site Ranking vs. NW-IBI for LWD assessments.

Figure 3.14. Saginaw River @ Zilwaukee (sg_zil01) riparian view. This site scored lowest in the composite NW-IBI, and was classified as "poor" by both types of assessment. This image is presented in color.

Figure 3.15. Grand River @ Ionia (gr_ion01, gr_ion02). This site was sampled in both 2001 and 2002, receiving a score of "fair" each time (composite assessment). This image is presented in color.

Figure 3.16. Manistee River @ High Bridge (ma_hbr). This site was scored as "good" in both 2001 and 2002 (composite assessment). Inset: Note clear water and slightly embedded coarse substrate. This image is presented in color.

Figure 3.17. Manistee River @ Coates Rd (ma_cts02). This site scored "excellent" in both assessment types. This image is presented in color.

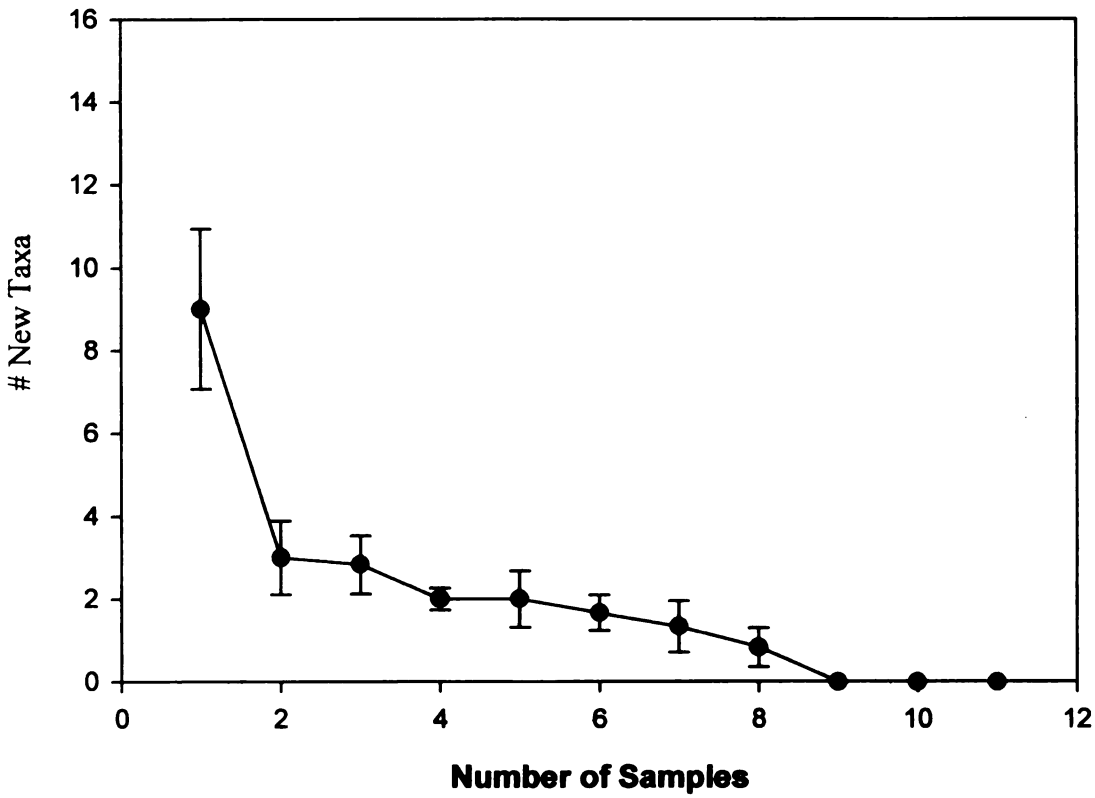


Figure 3.18. Mean number of new taxa collected in successive LWD samples. Note that after 8 samples, there were never new taxa collected, indicating that 8 samples should be sufficient for LWD assessments. These data were tabulated from 6 randomly chosen sites.

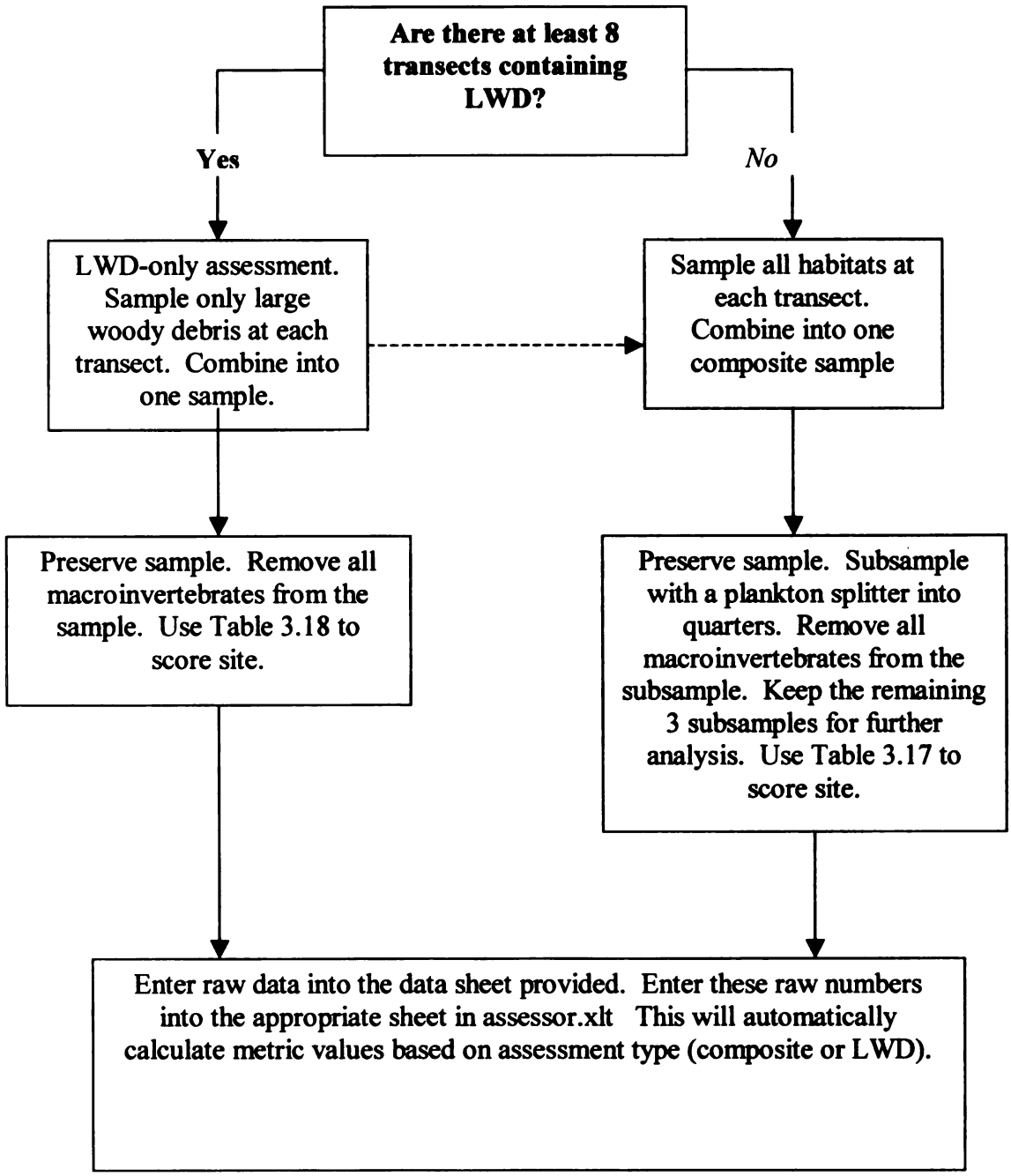


Figure 3.19. Flowchart illustrating the steps involved in the biological assessment of non-wadeable rivers in Michigan.

APPENDIX 1. COMPOSITE ASSESSMENT METRICS AND STRESSOR-RESPONSE RELATIONSHIPS

APPENDIX 1. COMPOSITE ASSESSMENT METRICS AND STRESSOR-RESPONSE RELATIONSHIPS

The following section provides more detailed information regarding specific metrics' response to various environmental parameters. For a summary of physical, chemical and landuse variables, see Tables 2.1 and 2.2. Only significant relationships are mentioned in this section. This information may be used when conducting composite assessments to diagnose specific human influences causing ecological impairment. See Table 3.19 for MLR results. In all cases the correlation coefficient was higher in backward elimination MLR (Table 3.19).

Composite Metric 1. Functional Feeding Group Diversity (FFG_DIV) (25 pts.). In addition to the parameters listed below, FFG diversity also shows a general response to riparian landuse (Table 3.19).

- MLR Forward Stepwise:
    1. Turbidity (-)
    2. Percent natural riparian landuse (+)
- MLR Backward Elimination
    1. Total Phosphorous (-)
    2. Conductivity
    3. Turbidity (-)
    4. Suspended chlorophyll

Composite Metric 2. Habitat Stability FFG Surrogate (HAB_STAB) [(# Scrapers+#Coll-Filt)/(#Coll-Gath+#Shredders)] (25 pts.):

- MLR Forward Stepwise:
    1. Conductivity
    2. Percent agricultural riparian landuse (+)
- MLR Backward Elimination

1. Percent urban landuse in the watershed (-)
2. Percent agricultural landuse in the watershed (-)
3. Percent natural landuse in the watershed (+)
4. Percent agricultural riparian landuse (-)

Composite Metric 3. Percent Trichoptera Abundance (PER_T) (20 pts.)

- MLR Forward Stepwise:

    1. Percent agricultural riparian landuse

- MLR Backward Elimination

    1. Percent agricultural riparian landuse

Composite Metric 4. EPT Richness (EPT_RICH) (8 pts.)

- MLR Forward Stepwise:

    1. Amount of LWD in the study reach
    2. Percent urban landuse in the watershed

- MLR Backward Elimination

    1. Amount of LWD in the study reach
    2. Percent urban landuse in the watershed

Composite Metric 5. Total Taxonomic Richness (RICH) (7 pts.)

- MLR Forward Stepwise:

    1. Percent urban landuse in the watershed

- MLR Backward Elimination: There were no significant relationships.

Composite Metric 6. Diptera Taxa Richness (DIP_RICH) (5 pts.)

- MLR Forward Stepwise:

    1. Total nitrogen

- MLR Backward Elimination

    1. Total nitrogen
    2. pH
    3. Turbidity
    4. Suspended chlorophyll

Composite Metric 7. Plecoptera Taxa Richness (P_RICH) (5 pts.)

- MLR Forward Stepwise:

    1. Amount of LWD in the study reach

- MLR Backward Elimination

    1. Amount of LWD in the study reach

Composite Metric 8. Percent Dominance (PER_DOM) (5 pts.). In addition to the parameters listed below, PER_DOM also showed significant correlations with riparian and watershed landuse.

- MLR Forward Stepwise:

    1. Percent natural riparian landuse

- MLR Backward Elimination

    1. Total phosphorous
    2. Conductivity

APPENDIX 2. LARGE WOODY DEBRIS (LWD) ASSESSMENT METRICS AND STRESSOR-RESPONSE RELATIONSHIPS

APPENDIX 2. LARGE WOODY DEBRIS (LWD) ASSESSMENT METRICS AND STRESSOR-RESPONSE RELATIONSHIPS

The following section provides more detailed information regarding specific metrics' response to various environmental parameters. For a summary of physical, chemical and landuse variables, see Tables 2.1 and 2.2. Only significant relationships are mentioned in this section. This information may be used when conducting LWD assessments to diagnose specific human influences causing ecological impairment. A complete list of environmental parameters included in MLR automatic stepwise regression can be examined in Tables 2.1 and 2.2. In all cases, correlation coefficients were equal or higher in the backward elimination method (Table 3.20).

LWD Metric 1. Habitat Stability FFG Surrogate (HAB_STAB) [(# Scrapers+#Coll-Filt)/(#Coll-Gath+#Shredders)] (20 pts.):

- MLR Forward Stepwise:

    3. Percent natural landuse in the watershed

- MLR Backward Elimination

    1. Total Nitrogen
    2. Percent agricultural riparian landuse
    3. Percent natural riparian landuse

LWD Metric 2. Functional Feeding Group Diversity (FFG_DIV) (20 pts.).

- MLR Forward Stepwise

    1. pH
    2. Percent urban watershed-wide landuse

- MLR Backward Elimination

    1. Total nitrogen
    2. Total phosphorous
    3. pH
    4. Percent natural riparian landuse

LWD Metric 3. Percent Dominant Taxon (PER_DOM) (20 pts.).

- MLR Forward Stepwise

    1. Dissolve oxygen
    2. Percent urban landuse in the watershed
    3. Percent natural riparian landuse

- MLR Backward Elimination

    1. Dissolve oxygen
    2. Percent urban landuse in the watershed
    3. Percent natural riparian landuse

LWD Metric 4. Percent Trichoptera Abundance (PER_T) (15 pts).

- MLR Forward Stepwise

    1. pH
    2. Percent agricultural landuse in the watershed
    3. Percent agricultural riparian landuse
- MLR Backward Elimination

    1. pH
    2. Percent urban landuse in the watershed
    3. Percent agricultural riparian landuse

LWD Metric 5. Diptera Taxa Richness (DIP_RICH) (15 pts.).

- MLR Forward Stepwise

    1. Total nitrogen
    2. Percent urban landuse in the watershed
    3. Percent natural landuse in the watershed
    4. Percent urban riparian landuse
- MLR Backward Elimination

    1. Total nitrogen
    2. Percent urban landuse in the watershed
    3. Percent natural landuse in the watershed
    4. Percent urban riparian landuse

LWD Metric 6. Percent Scrapers (SCR) (10 pts.). In addition to the parameters listed below, this metric has a broad response to riparian landuse.

- MLR Forward Stepwise
    1. Percent natural landuse in the watershed
- MLR Backward Elimination
    1. Total phosphorous
    2. Conductivity
    3. pH

APPENDIX 3:

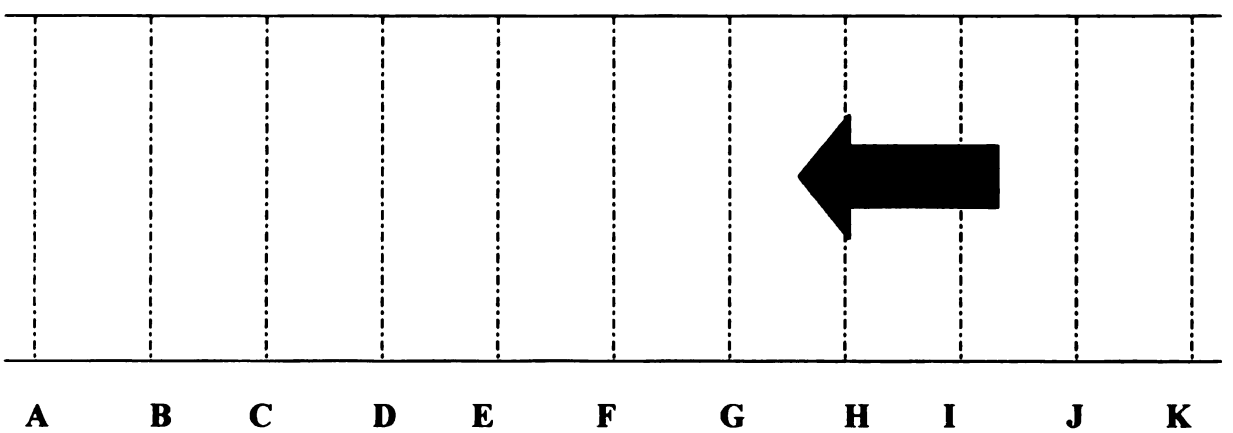
FIELD MANUAL FOR THE BIOLOGICAL ASSESSMENT OF NON-WADEABLE RIVERS IN MICHIGAN

I. GENERAL PROCEDURE

A. Use the equipment checklist to ensure all necessary equipment is brought along for the assessment.

B. Locate the reach of interest. Assessment of non-wadeable rivers will be at the reach scale. However, test reaches should be representative of the larger river and catchment. Considerations of which reaches to evaluate include:

1. Proximity to urban centers (e.g., downstream from a metropolitan area or intensively farmed area).
2. Ease of access. Can the crew get to the site with the needed equipment?
3. Specific stressors known to affect a certain area.
4. Motor to the downstream end of the study reach and mark this area. This is Transect A. Additional transects are located ≈ 200m upstream of each subsequent transect. This should be done by the Habitat Assessment Crew. Given a total reach size of 2000m, there are 11 total transect (A-K).
5. Determine randomly (e.g., flipping a coin) which bank to sample.
6. Sample all available habitats (or just the woody debris) within ≈ 10m upstream and downstream of the transect. Transects should be marked with flagging and labeled (A-K) by the Habitat Crew in case they need to be relocated at later times during the field portion of the assessment. Sampling should take place in shoreline areas (<1m deep). See the next section for detailed description of sampling procedures.
7. Using the taxonomic data sheet, record all taxa in the sample and the abundance of each. This may need to be done at the lab for composite samples. LWD samples may be processed in the field by experienced field technicians.



Schematic diagram of a non-wadeable study reach. Total length is 2000m. Transects are labeled A-K and are evenly spaced 200m apart. Macroinvertebrate sampling takes place at each transect on randomly-chosen banks. Arrow indicates the direction of flow.

II. DETAILED SAMPLING PROCEDURES

A. Composite Samples: Use this method if large woody debris is not present or for a more detailed assessment of the reach. Using this approach, the biological assessment will reflect the available habitat as well as in-stream water quality. This sampling procedure involves sampling all available habitats at each transect and combining the individual samples into one composite for the entire reach. At each transect:

1. Tally the individual habitat types. These include:
    a) Fine particulate organic matter (FPOM)
    b) Sand
    c) Gravel
    d) Cobble
    e) Large woody debris (LWD)
    f) Macrophytes
2. For each habitat type, take timed samples (15 seconds each) with a D-frame aquatic dip net with mesh size = 0.5mm. Habitat-specific considerations are as follows:

    a) FPOM: If there is flow through the sampling area, use kick methods to reduce the amount of detritus in the sample. If there is no flow, sweep the net along the bottom and make sure to wash as much detritus from the net as possible before preserving the sample.

    b) Sand: Same as above.

    c) Gravel: Same as above.

    d) Cobble: It is difficult to take timed sweeps of cobble habitat. Therefore, try to choose a piece of cobble at least 15 cm in diameter. Place the cobble in a bucket and brush organisms off with a toilet brush.

    e) Large Woody Debris (LWD): Sampling LWD presents challenges, especially when the debris cannot be removed from the river. We suggest using a toilet brush to dislodge organisms from the LWD and following closely behind with the net. If there is high flow in the area begin sampled, make sure the net opens into the current and the brush is 'upstream' of the net. Do this for ≈ 15 seconds.

    f) Macrophytes: If there are macrophytes in the study reach, take timed sweeps (≈ 15 seconds) of the stems to dislodge attached macroinvertebrates.
3. Empty the net into a white enamel pan filled with water. This allows you to easily wash out the net (you may need to pick attached organisms from the net with forceps).
4. Remove as much detritus as possible before pouring the sample into a 500 µm sieve to remove excess water.
5. Place each individual sample for each habitat at each transect into a bucket and preserve in the field with 95% EtOH. Further processing will be done in the laboratory.

B. Habitat-Specific Sampling: If the study site contains sufficient amounts of LWD, you may evaluate the reach by sampling only LWD habitats. This will significantly reduce sample processing time and allow an evaluation of the reach's ecological integrity that is more independent of the habitat assessment. Follow the procedures above to sample LWD. Because of the inherent variability in non-wadeable systems, this should only be done if there is sufficient LWD habitat (e.g., the number of transects with LWD habitat is ≥ 8).

C. Further Sample Processing
   1. Composite Samples will be returned to the laboratory, subsampled (quarters), and macroinvertebrates will be sorted and identified to family level.
   2. LWD Samples may be sorted and identified in the field. If this is done, make sure to enter raw data into the Bioassessment Field Data Sheet (back page).

III. QUALITY ASSURANCE/QUALITY CONTROL

A. For any macroinvertebrate identifications you are not sure about, place representative specimens in vials containing preservative. This will be especially important if you are sorting and counting invertebrate samples in the field, such as when doing LWD habitat specific assessments.

B. Clearly label each specimen with site information and number of each 'type' in the sample.

C. Take the specimens back to the laboratory for examination under a microscope.

D. For composite sample assessments, return one of the subsamples (1/4) to the laboratory for storage. This will allow reassessment at a later time and comparison of subsamples.

EQUIPMENT CHECKLIST FOR NON-WADEABLE RIVER BIOASSESSMENT

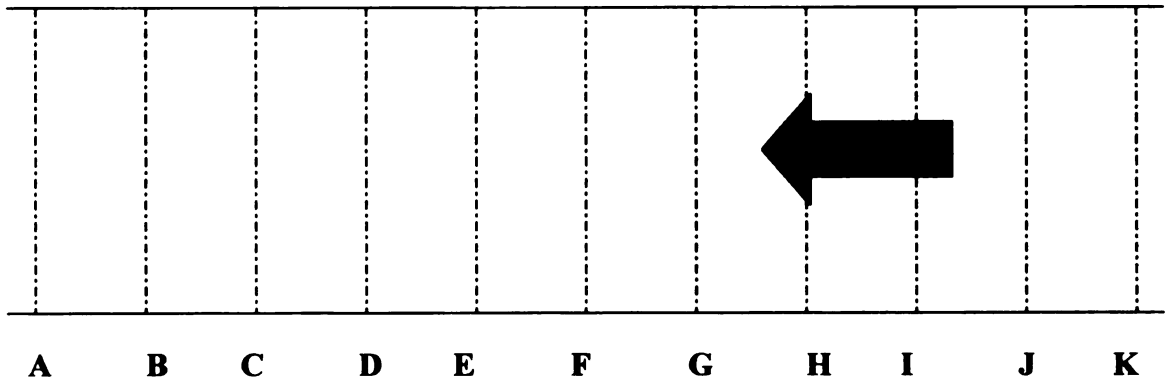
The following items should be included with the crew responsible for the collection of macroinvertebrates:

√	QNTY	ITEM
	2	500 µm mesh D-frame aquatic dip nets (2)
		5L 95% Ethanol
	2	Standard toilet brushes (2)
	1	Large bucket with lid for samples
	2	Forceps (2) for picking organisms from D-frame net.
	2	500um sieves (2) for processing samples.
	2	White enamel pans (2) for sorting organisms
	10-20	Vials for voucher specimens (If sample processing is to be done in the field)
		Labels for voucher specimens
		Color identification plates
		Non-wadeable biological assessment data sheet (front and back)
	1	Plankton splitter (If sample processing is to be done in the field)
	2	Magnifying lenses (2) (If sample processing is to be done in the field)

BIOASSESSMENT FIELD DATA SHEET
(Front page)

DATE:		**CREW**		
RIVER:		**REACH LOCATION**		
		GPS or Gazetteer Info	**Other information**	
			Upstream **Downstream**	**Of (City, Dam, etc.)**
Assessment Type:	**COMPOSITE** **LWD**	**Other Notes:**		

On the diagram below, mark the locations at which macroinvertebrate samples were taken.



For composite assessments, note which macroinvertebrate habitats were present at each transect.

A	**F**	**Sa**	**C**	**Cb**	**W**	**M**	**G**	**F**	**Sa**	**C**	**Cb**	**W**	**M**
B	**F**	**Sa**	**C**	**Cb**	**W**	**M**	**H**	**F**	**Sa**	**C**	**Cb**	**W**	**M**
C	**F**	**Sa**	**C**	**Cb**	**W**	**M**	**I**	**F**	**Sa**	**C**	**Cb**	**W**	**M**
D	**F**	**Sa**	**C**	**Cb**	**W**	**M**	**J**	**F**	**Sa**	**C**	**Cb**	**W**	**M**
E	**F**	**Sa**	**C**	**Cb**	**W**	**M**	**K**	**F**	**Sa**	**C**	**Cb**	**W**	**M**
F	**F**	**Sa**	**C**	**Cb**	**W**	**M**	**Total Samples:**						

F=FPOM; Sa=Sand; C=Coarse substrates; Cb=Cobble; W=Large Woody Debris; M=Macrophytes

BIOASSESSMENT FIELD DATA SHEET
(Back page)

SAMPLE TYPE	COMPOSITE LWD	This data sheet allows you to quickly summarize your field data. Box numbers correspond to the MS Excel file (assessor.xlt) used for metric scoring and summary of ecological condition. When entered correctly, scores are automatically calculated for each site.	
		Enter these values into corresponding box in Excel template (assessor.xlt).	Box Number
Total Abundance			1
Total Richness			2
Number of Ephemeroptera Families			3
Number of Plecoptera Families			4
Number of Trichoptera Families			5
Number of Diptera Taxa			6
Trichoptera Abundance			7
Abundance of Dominant Taxon			8
Shredder Abundance			9
Scraper Abundance			10
Coll-Filterer Abundance			11
Coll-Gath Abundance			12
Predator Abundance			13

APPENDIX 4

Record of Deposition of Voucher Specimens*

The specimens listed on the following sheet(s) have been deposited in the named museum(s) as samples of those species or other taxa, which were used in this research. Voucher recognition labels bearing the Voucher No. have been attached or included in fluid-preserved specimens.

Voucher No.: 2004-08

Title of thesis or dissertation (or other research projects):

BIOLOGICAL EVALUATION OF NON-WADEABLE RIVERS IN MICHIGAN

Museum(s) where deposited and abbreviations for table on following sheets:

Entomology Museum, Michigan State University (MSU)

Other Museums:

Investigator's Name(s) (typed)
Kelly James Wessell

Date 12-16-2004

*Reference: Yoshimoto, C. M. 1978. Voucher Specimens for Entomology in North America. Bull. Entomol. Soc. Amer. 24: 141-42.

Deposit as follows:

Original: Include as Appendix 1 in ribbon copy of thesis or dissertation.

Copies: Include as Appendix 1 in copies of thesis or dissertation.
Museum(s) files.
Research project files.

This form is available from and the Voucher No. is assigned by the Curator, Michigan State University Entomology Museum.

Appendix 4

Voucher Specimen Data

Page 1 of 12 Pages

Species or other taxon			Label data for specimens collected or used and deposited	Number of: Eggs	Larvae	Nymphs	Pupae	Adults ♀	Adults ♂	Other	Museum where deposited
Athericidae	*Atherix*	sp.	01-01 MI Newago Co Muskegon R		1						MSU
Ceratopogonidae	*Atrichopogon*	sp.	01-02 MI Monroe Co Raisin R		1						MSU
Ceratopogonidae	*Bezzia*	sp.	01-03 MI Monroe Co Raisin R		1						MSU
Ceratopogonidae	*Ceratopogon*	sp.	01-04 MI Kent Co Grand R		1						MSU
Ceratopogonidae	*Probezzia*	sp.	01-05 MI Saginaw Co Titabaw. R.		1						MSU
Ceratopogonidae	*Sphaeromias*	sp.	01-06 MI Monroe Co Raisin R		1						MSU
Chaoboridae	*Chaoborus*	sp.	01-07 MI Manistee Co. Manistee R.		1						MSU
Chironomidae			01-08 MI Mecosta Co Muskegon R		1						MSU
Chironomidae			01-09 MI Kent Co. Grand R.				1				MSU
Culicidae	*Anopheles*	sp.	01-10 MI Manistee Co. Manistee R.		1						MSU
Dolichopodidae	*Hydrophorus*	sp.	01-11 MI Manistee Co. Manistee R.		1						MSU
Dolichopodidae	*Rhaphium*	sp.	01-12 MI Saginaw Co. Saginaw R.		1						MSU
Empididae	*Hemerodromia*	sp.	01-13 MI Kent Co. Grand R.		1						MSU
Empididae	*Hemerodromia*	sp.	01-14 MI Manistee Co. Manistee R.				1				MSU
Ephydridae	*Discocerina*	sp.	01-15 MI Kent Co. Grand R.		1						MSU
Ephydridae	*Hydrellia*	sp.	01-16 MI Mecosta Co Muskegon R		1						MSU
Muscidae			01-17 MI Kent Co. Grand R.		1						MSU

(Use additional sheets if necessary)

Investigator's Name(s) (typed)

Kelly James Wessell

Todd White

Date 12/15/04

Voucher No 2004-08

Received the above listed specimens for deposit in the Michigan State University Entomology Museum.

Curator

17 Dec 2004

Date

Appendix 4

Voucher Specimen Data

Page 2 of 12 Pages

Species or other taxon			Label data for specimens collected or used and deposited	Number of: Eggs	Larvae	Nymphs	Pupae	Adults ♀	Adults ♂	Other	Museum where deposited
Simuliidae	*Simulium*	sp.	01-18 MI Kent Co Grand R.		1						MSU
Simuliidae	*Simulium*	sp.	01-19 MI Newago Co. Muskegon R.				1				MSU
Stratiomyidae	*Stratiomys*	sp.	01-20 MI Iosco Co. AuSable R.		1						MSU
Tabanidae	*Chrysops*	sp.	01-21 MI Manistee Co. Manistee R.		1						MSU
Tipulidae	*Antocha*	sp.	01-22 MI Manistee Co. Manistee R.		1						MSU
Tipulidae	*Antocha*	sp.	01-23 MI Manistee Co. Manistee R.		1						MSU
Tipulidae	*Limonia*	sp.	01-24 MI Berrien Co. St. Joseph R.		1						MSU
Tipulidae	*Ormosia*	sp.	01-25 MI Manistee Co. Manistee R.		1						MSU
Tipulidae	*Tipula*	sp.	01-26 MI Monroe Co. Raisin R.		1						MSU
Tipulidae	*Tipula*	sp.	01-27 MI Iosco Co. AuSable R.				1				MSU
Baetiscidae	*Baetisca*	sp.	02-01 MI Menominee Co. Menominee R		1						MSU
Caenidae	*Brachycercus*	sp.	02-02 MI Manistee Co. Manistee R.		1						MSU
Caenidae	*Caenis*	sp.	02-03 MI Monroe Co. Raisin R.		1						MSU
Caenidae	*Cercobrachys*	sp.	02-04 MI Manistee Co. Manistee R.		1						MSU
Ephemerellidae	*Drunella*	sp.	02-05 MI Newago Co. Muskegon R.		1						MSU
Ephemerellidae	*Eurylophella*	sp.	02-06 MI Mecosta Co. Muskegon R.		1						MSU
Ephemerellidae	*Seratella*	sp.	02-07 MI Muskegon C. Muskegon R.		1						MSU

(Use additional sheets if necessary)

Investigator's Name(s) (typed) Voucher No

Kelly James Wessell

Todd White

Received the above listed specimens for deposit in the Michigan State University Entomology Museum.

Date 12/15/04

Curator Date

Appendix 4

Voucher Specimen Data

Page_3_of_12_Pages

Species or other taxon		Label data for specimens collected or used and deposited	Number of:						Museum where deposited
			Eggs	Larvae	Nymphs	Pupae	Adults ♀	Adults ♂	Other	
Ephemerellidae	*Timpanoga* sp.	02-08 MI Ionia Co Grand R.		1						MSU
Ephemeridae	*Ephemera* sp.	02-09 MI Kent Co. Grand R.		1						MSU
Ephemeridae	*Hexagenia* sp.	02-10 MI Saginaw Co. Saginaw R.		1						MSU
Heptageniidae	*Heptagenia* sp.	02-11 MI Monroe Co. Raisin R.		1						MSU
Heptageniidae	*Leucrocuta* sp.	02-12 MI Kent Co. Grand R.		1						MSU
Heptageniidae	*Rhithrogena* sp.	02-13 MI Newago Co. Muskegon R.		1						MSU
Heptageniidae	*Stenacron* sp.	02-14 MI Kent Co. Grand R.		1						MSU
Heptageniidae	*Stenonema* sp.	02-15 MI Muskegon Co. Muskegon R		1						MSU
Isonychiidae	*Isonychia* sp.	02-16 MI Kent Co. Grand R.		1						MSU
Leptophlebiidae	*Leptophlebia* sp.	02-17 MI Menominee Co. Menominee R		1						MSU
Polymitarcyidae	*Ephoron* sp.	02-18 IN Elkhart Co. St Joseph R.		1						MSU
Potamanthidae	*Anthopotamus* sp.	02-19 MI Kent Co. Grand R.		1						MSU
Tricorythidae	*Tricorythodes* sp.	02-20 MI Kent Co. Grand R.		1						MSU
Baetidae		02-21 MI Newago Co. Muskegon R		1						MSU
Leptophlebiidae	*Chorotterpes* sp.	02-22 MI Menominee Co. Menominee R.		1						MSU
Carabidae		03-01 MI Saginaw Co. Shiawassee R.		1						MSU
Chrysomelidae	*Donacia* sp.	03-02 MI Saginaw Co. Shiawassee R.						1		MSU

(Use additional sheets if necessary)

Investigator's Name(s) (typed)

Kelly James Wessell

Todd White

Date 12/15/04

Voucher No.

Received the above listed specimens for deposit in the Michigan State University Entomology Museum.

Curator **Date**

Appendix 4

Voucher Specimen Data

Page 4 of 12 Pages

Species or other taxon		Label data for specimens collected or used and deposited	Number of:						
			Eggs	Larvae	Nymphs	Pupae	Adults ♀	Adults ♂	Other	Museum where deposite d
Curculionidae		03-03 MI Ionia Co. Grand R.					1			MSU
Dytiscidae	*Copelatus* sp.	03-04 MI Manistee Co. Manistee R.		1						MSU
Dytiscidae	*Coptotomus* sp.	03-05 MI Luce Co. Tahquanmenon R.		1						MSU
Dytiscidae	*Coptotomus* sp.	03-06 MI Luce Co. Tahquanmenon R.						1		MSU
Dytiscidae	*Hydroporus* sp.	03-07 MI Chippewa Co. Tahquamenon R.						1		MSU
Dytiscidae	*Hydrovatus* sp.	03-08 MI Iosco Co. AuSable R.						1		MSU
Dytiscidae	*Laccophilus* sp.	03-09 MI Saginaw Co. Shiawassee R.			1					MSU
Dytiscidae	*Laccophilus* sp.	03-10 MI Manistee Co. Manistee R.						1		MSU
Dytiscidae	*Nebrioporus/Stict* sp.	03-11 MI Manistee Co. Manistee R.						1		MSU
Elmidae	*Ancyronyx* sp.	03-12 MI Manistee Co. Manistee R.			1					MSU
Elmidae	*Ancyronyx* sp.	03-13 MI Manistee Co. Manistee R.					1			MSU
Elmidae	*Dubraphia* sp.	03-14 MI Manistee Co. Manistee R.			1					MSU
Elmidae	*Dubraphia* sp.	03-15 MI Monroe Co. Raisin R.					1			MSU
Elmidae	*Macronychus* sp.	03-16 MI Kent Co. Grand R.			1					MSU
Elmidae	*Macronychus* sp.	03-17 MI Manistee Co. Manistee R.					1			MSU
Elmidae	*Optioservus* sp.	03-18 MI Newago Co. Muskegon R.			1					MSU
Elmidae	*Optioservus* sp.	03-19 MI Iosco Co. AuSable R.					1			MSU

(Use additional sheets if necessary)

Investigator's Name(s) (typed)

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Todd White

Date 12/15/04

Voucher No.

Received the above listed specimens for deposit in the Michigan State University Entomology Museum.

Curator Date

Appendix 4

Voucher Specimen Data

Page 5 of 12 Pages

Species or other taxon			Label data for specimens collected or used and deposited	Number of:						Museum where deposited
				Eggs	Larvae	Nymphs	Pupae	Adults ♀	Adults ♂	Other	
Elmidae	Stenelmis	sp.	03-20 MI Kent Co. Grand R.		1						MSU
Elmidae	Stenelmis	sp.	03-21 MI Kent Co. Grand R.					1			MSU
Gyrinidae	Dineutus	sp.	03-22 MI Ionia Co. Grand R.					1			MSU
Gyrinidae	Dineutus	sp.	03-23 MI Manistee Co. Manistee R.			1					MSU
Gyrinidae	Gyrinus	sp.	03-24 MI Kalamazoo Co. Kalamazoo R					1			MSU
Gyrinidae	Gyrinus	sp.	03-25 MI Iosco Co. AuSable R.			1					MSU
Haliplidae	Haliplus	sp.	03-26 MI Luce Co. Tahquamenon R.					1			MSU
Haliplidae	Haliplus	sp.	03-27 MI Kalamazoo Co. Kalamazoo R			1					MSU
Haliplidae	Peltodytes	sp.	03-28 MI Ionia Co. Grand R.					1			MSU
Haliplidae	Peltodytes	sp.	03-29 MI Manistee Co. Manistee R.					1			MSU
Hydrophilidae	Berosus	sp.	03-30 MI Saginaw Co. Saginaw R.					1			MSU
Hydrophilidae	Berosus	sp.	03-31 MI Iosco Co. AuSable R.			1					MSU
Hydrophilidae	Enochrus	sp.	03-32 MI Kent Co. Grand R.					1			MSU
Hydrophilidae	Laccobius	sp.	03-33 MI Manistee Co. Manistee R.					1			MSU
Hydrophilidae	Laccobius	sp.	03-34 MI Iosco Co. AuSable R.			1					MSU
Hydrophilidae	Sperchopsis	sp.	03-35 MI Kent Co. Grand R.						1		MSU
Hydrophilidae	Tropisternus	sp.	03-36 MI Newago Co. Muskegon R.					1			MSU

(Use additional sheets if necessary)

Investigator's Name(s) (typed)

Kelly James Wessell

Todd White

Date 12/15/04

Voucher No.

Received the above listed specimens for deposit in the Michigan State University Entomology Museum.

Curator Date

Appendix 4

Voucher Specimen Data

Page 6 of 12 Pages

Species or other taxon			Label data for specimens collected or used and deposited	Number of:						Museum where deposited
				Eggs	Larvae	Nymphs	Pupae	Adults ♀	Adults ♂	Other	
Hydrophilidae	*Tropisternus*	sp.	03-36 MI Newago Co Muskegon R.						1		MSU
Hydrophilidae	*Tropisternus*	sp.	03-37 MI Kent Co. Grand R.								MSU
Noteridae	*Hydrocanthus*	sp.	03-38 MI Kent Co. Grand R.					1			MSU
Psephenidae	*Ectopria*	sp.	03-39 MI Manistee Co. Manistee R.		1						MSU
Psephenidae	*Psephenus*	sp.	03-40 MI Newago Co. Muskegon R.		1						MSU
Scirtidae	*Scirtes*	sp.	03-41 MI Ionia Co. Grand R.		1						MSU
Belostomatidae	*Belostomata*	sp.	04-01 MI Luce Co. Tahquamenon R.				1				MSU
			04-02 MI Saginaw Co. Shiawassee R.				1				MSU
Corixidae			04-03 MI Kalamazoo Co. Kalamazoo R.				1				MSU
Gerridae	*Aquarius*	sp.	04-04 MI Menominee Co. Menominee R.				1				MSU
Gerridae	*Limnoporus*	sp.	04-05 MI Kent Co. Grand R.				1				MSU
Gerridae	*Metrobates*	sp.	04-06 MI Manistee Co. Manistee R.				1				MSU
Gerridae	*Neogerris*	sp.	04-07 MI Saginaw Co. Tittabawassee R.				1				MSU
Gerridae	*Rheumatobates*	sp.	04-08 MI Newago Co. Muskegon R.				1				MSU
Gerridae	*Trepobates*	sp.	04-09 MI Saginaw Co. Shiawassee R.				1				MSU
Hebridae	*Hebrus*	sp.	04-10 MI Ionia Co. Grand R.				1				MSU
Mesoveliidae	*Mesovelia*	sp.	04-11 MI Luce Co. Tahquamenon R.				1				MSU
Nepidae	*Ranatra*	sp.									

(Use additional sheets if necessary)

Investigator's Name(s) (typed)

Kelly James Wessell

Todd White

Date 12/15/04

Voucher No.

Received the above listed specimens for deposit in the Michigan State University Entomology Museum.

Curator Date

Appendix 4

Voucher Specimen Data

Page 7 of 12 Pages

Species or other taxon			Label data for specimens collected or used and deposited	Number of:						
				Eggs	Larvae	Nymphs	Pupae	Adults ♀	Adults ♂	Other	Museum where deposited
Notonectidae	*Buenoa*	sp.	04-12 MI Manistee Co. Manistee R.					1			MSU
Pleidae	*Neoplea*	sp.	04-13 MI Kent Co. Grand R.					1			MSU
Saldidae	*Saldus*	sp.	04-14 MI Saginaw Co. Shiawassee R.					1			MSU
Veliidae	*Microvelia*	sp.	04-15 MI Muskegon Co. Muskegon R					1			MSU
Veliidae	*Rhagovelia*	sp.	04-16 MI Kalamazoo Co. Kalamazoo R					1			MSU
Mymaridae	*Caraphractus*	sp.	05-01 MI Manistee Co. Manistee R.		1						MSU
Nepticulidae	*Nepticula*	sp.	06-01 MI Manistee Co. Manistee R.		1						MSU
Pyralidae	*Acentria*	sp.	06-02 MI Iosco Co. AuSable R.		1						MSU
Pyralidae	*Paraponyx*	sp.	06-03 MI Luce Co. Tahquamenon R.		1						MSU
Pyralidae	*Petrophila*	sp.	06-04 MI Iosco Co. AuSable R.		1						MSU
Corydalidae	*Chauliodes*	sp.	07-01 MI Berrien Co. St. Joseph R.		1						MSU
Sialidae	*Sialis*	sp.	07-02 MI Newago Co. Muskegon R.		1						MSU
Sisyridae	*Climacia*	sp.	08-01 MI Muskegon Co. Muskegon R.		1						MSU
Sisyridae	*Sisyra*	sp.	08-02 MI Kent Co. Grand R.		1						MSU
Aeshnidae	*Anax*	sp.	09-01 MI Luce Co. Tahquamenon R.			1					MSU
Aeshnidae	*Boyeria*	sp.	09-02 MI Elkhart Co. St. Joseph R			1					MSU
Cordulidae	*Cordulia*	sp.	09-03 Manistee Co. Manistee R.			1					MSU

(Use additional sheets if necessary)

Investigator's Name(s) (typed)

Kelly James Wessell

Todd White

Date 12/15/04

Voucher No

Received the above listed specimens for deposit in the Michigan State University Entomology Museum.

Curator Date

Appendix 4

Voucher Specimen Data

Page 8 of 12 Pages

Species or other taxon			Label data for specimens collected or used and deposited	Number of:						Museum where deposited
				Eggs	Larvae	Nymphs	Pupae	Adults ♀	Adults ♂	Other	
Cordulliidae	*Macromia*	sp.	09-04 MI Menominee Co. Menominee R.			1				MSU
Cordulliidae	*Neurocordulia*	sp.	09-05 MI Iosco Co. AuSable R.			1				MSU
Gomphidae	*Gomphus*	sp.	09-06 MI Manistee Co Manistee R.			1				MSU
Gomphidae	*Stylurus*	sp.	09-07 MI Muskegon Co. Muskegon R			1				MSU
Libellulidae	*Sympetrum*	sp.	09-08 MI Luce Co. Tahquamenon R			1				MSU
Calopterygidae	*Calopteryx*	sp.	09-09 MI Luce Co. Tahquamenon R.			1				MSU
Calopterygidae	*Hetaerina*	sp.	09-10 MI Kalamazoo Co. Kalamazoo R.			1				MSU
Coenagrionidae	*Argia*	sp.	09-11 MI Kalamazoo Co. Kalamazoo R.			1				MSU
Coenagrionidae	*Coenagrion/Enall*	sp.	09-12 MI Muskegon Co. Muskegon R.			1				MSU
Coenagrionidae	*Ischnura*	sp.	09-13 MI Iosco Co. AuSable R.			1				MSU
Lestidae	*Lestes*	sp.	09-14 MI Kalamazoo Co. Kalamazoo R			1				MSU
Perlidae	*Acroneuria*	sp.	10-01 MI Newago Co. Muskegon R.			1				MSU
Perlidae	*Neoperla*	sp.	10-02 IN Elkhart Co. St. Joseph R.			1				MSU
Perlidae	*Paragnetina*	sp.	10-03 MI Muskegon Co. Muskegon R.			1				MSU
Perlidae	*Perlesta*	sp.	10-04 MI Muskegon Co. Muskegon R.			1				MSU
Pteronarcyidae	*Pteronarcys*	sp.	10-05 MI Muskegon Co. Muskegon R.			1				MSU
Brachycentridae	*Brachycentrus*	sp.	11-01 MI Manistee Co. Manistee R.		1					MSU

(Use additional sheets if necessary)

Investigator's Name(s) (typed)

Kelly James Wessell

Todd White

Date 12/15/04

Voucher No.

Received the above listed specimens for deposit in the Michigan State University Entomology Museum.

Curator Date

Appendix 4

Voucher Specimen Data

Page 9 of 12 Pages

Species or other taxon			Label data for specimens collected or used and deposited	Number of:						Museum where deposited
				Eggs	Larvae	Nymphs	Pupae	Adults ♀	Adults ♂	Other	
Brachycentridae	*Micrasema*	sp.	11-02 MI Newago Co. Muskegon R.		1						MSU
Dipseudopsidae	*Phylocentropus*	sp.	11-03 MI Luce Co. Tahquamenon R.		1						MSU
Glossosomatidae	*Protoptila*	sp.	11-04 MI Manistee Co. Manistee R.		1						MSU
Glossosomatidae	*Protoptila*	sp.	11-05 MI Manistee Co. Manistee R.		1						MSU
Helicopsychidae	*Helicopsyche*	sp.	11-06 MI Manistee Co. Manistee R.		1						MSU
Helicopsychidae	*Helicopsyche*	sp.	11-07 MI Newago Co. Muskegon R.				1				MSU
Hydropsychidae	*Cheumatopsyche*	sp.	11-08 MI Kent Co. Grand R.			1					MSU
Hydropsychidae	*Hydropsyche*	sp.	11-09 MI Kent Co. Grand R.			1					MSU
Hydropsychidae	*Macrostemum*	sp.	11-10 MI Berrien Co. St. Joseph R.			1					MSU
Hydropsychidae	*Potamyia*	sp.	11-11 MI Kent Co. Grand R.			1					MSU
Hydropsychidae			11-12 MI Muskegon Co. Muskegon R.			1	1				MSU
Hydroptilidae	*Agraylea*	sp.	11-13 MI Iosco Co. AuSable R.			1					MSU
Hydroptilidae	*Hydroptila*	sp.	11-14 MI Newago Co. Muskegon R.			1					MSU
Hydroptilidae	*Ithytricia*	sp.	11-15 MI Chippewa Co. Tahquamenon R			1					MSU
Hydroptilidae	*Ochrotrichia*	sp.	11-16 MI Manistee Co. Manistee R			1					MSU
Hydroptilidae	*Orthotrichia*	sp.	11-17 MI Iosco Co. AuSable R.			1					MSU
Hydroptilidae	*Oxyethira*	sp.	11-18 MI Iosco Co. AuSable R.			1					MSU

(Use additional sheets if necessary)

Investigator's Name(s) (typed)

Kelly James Wessell

Todd White

Date 12/15/04

Voucher No._____

Received the above listed specimens for deposit in the Michigan State University Entomology Museum.

Curator _____ Date _____

Appendix 4

Voucher Specimen Data

Page_10_ of_ 12_Pages

Species or other taxon		Label data for specimens collected or used and deposited	Number of:					Museum where deposited
			Eggs	Larvae	Nymphs	Pupae	Adults ♀	Adults ♂	Other	
Hydroptilidae		11-19 MI Newago Co. Muskegon R.				1				MSU
Lepidostomatidae	*Lepidostoma* sp.	11-20 IN Elkhart Co. St. Joseph R.		1						MSU
Leptoceridae	*Ceraclea* sp.	11-21 MI Kent Co. Grand R.		1						MSU
Leptoceridae	*Ceraclea* sp.	11-22 MI Newago Co. Muskegon R				1				MSU
Leptoceridae	*Mystacides* sp.	11-23 MI Iosco Co. AuSable R.		1						MSU
Leptoceridae	*Nectopsyche* sp.	11-24 MI Newago Co. Muskegon R.		1						MSU
Leptoceridae	*Nectopsyche* sp.	11-25 MI Manistee Co. Manistee R.				1				MSU
Leptoceridae	*Oecetis* sp.	11-26 MI Manistee Co. Manistee R.		1						MSU
Leptoceridae	*Oecetis* sp.	11-27 MI Saginaw Co. Tittabawassee R.				1				MSU
Leptoceridae	*Setodes* sp.	11-28 MI Manistee Co. Manistee R.		1						MSU
Leptoceridae	*Ylodes* sp.	11-29 MI Iosco Co. AuSable R.		1						MSU
Limnephilidae	*Pycnopsyche* sp.	11-30 MI Newago Co. Muskegon R		1						MSU
Philopotamidae	*Chimarra* sp.	11-31 MI Newago Co. Muskegon R.		1						MSU
Philopotamidae	*Chimarra* sp.	11-32 MI Iosco Co. AuSable R.		1						MSU
Phryganeidae	*Phryganea* sp.	11-33 MI Luce Co. Tahquamenon R.		1						MSU
Polycentropodidae	*Cernotina* sp.	11-34 MI Iosco Co. AuSable R.		1						MSU
Polycentropodidae	*Cyrnellus* sp.	11-35 MI Kalamazoo Co. Kalamazoo R.		1						MSU

(Use additional sheets if necessary)

Investigator's Name(s) (typed)

Kelly James Wessell

Todd White

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Voucher No._____

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Curator Date

Appendix 4

Voucher Specimen Data

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Species or other taxon			Label data for specimens collected or used and deposited	Number of:						Museum where deposited
				Eggs	Larvae	Nymphs	Pupae	Adults ♀	Adults ♂	Other	
Polycentropodidae	*Neureclipsis*	sp.	11-36 MI Manistee Co. Manistee R.		1						MSU
Polycentropodidae	*Paranyctiophylax*	sp.	11-37 MI Mecosta Co. Muskegon R.		1						MSU
Polycentropodidae	*Polycentropus*	sp.	11-38 MI Luce Co. Tahquamenon R.		1						MSU
Polycentropodidae	*Pupae*		11-39 MI Mecosta Co. Muskegon R.				1				MSU
Psychomyiidae	*Lype*	sp.	11-40 MI Manistee Co. Manistee R.			1					MSU
Psychomyiidae	*Psychomyia*	sp.	11-41 MI Newago Co. Muskegon R.			1					MSU
Uenoidae	*Neophylax*	sp.	11-42 MI Newago Co. Muskegon R.			1					MSU
Arachnida	Hydrachnida		12-01 MI Iosco Co. AuSable R.								MSU
Annelida	Oligochaeta		12-02 MI Muskegon Co. Muskegon R.								MSU
Annelida	Hirudinea		12-03 MI Iosco Co. AuSable R.								MSU
Bryozoa			12-04 MI Kent Co. Grand R.								MSU
Crustacea	Amphipoda		12-05 MI Newago Co. Muskegon R.								MSU
Crustacea	Branchiura		12-06 MI Saginaw Co. Shiawassee R.								MSU
Crustacea	Isopoda		12-07 MI Iosco Co. AuSable R.								MSU
Crustacea	Decapoda		12-08 IN Elkhart Co. St. Joseph R.								MSU
Mollusca	Bivalvia		12-09 IN Elkhart Co. St. Joseph R.								MSU
Mollusca	Bivalvia		12-10 MI Monroe Co Raisin R.								MSU

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Voucher Specimen Data

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Species or other taxon		Label data for specimens collected or used and deposited	Number of:						Museum where deposited
			Eggs	Larvae	Nymphs	Pupae	Adults ♀	Adults ♂	Other	
Mollusca	Bivalvia	12-11 MI Manistee Co. Manistee R.								MSU
Mollusca	Gastropoda	12-12 MI Newago Co. Muskegon R.								MSU
Mollusca	Gastropoda	12-13 MI Newago Co. Muskegon R.								MSU
Mollusca	Gastropoda	12-14 MI Manistee Co. Manistee R.								MSU
Mollusca	Gastropoda	12-15 MI Saginaw Co. Shiawassee R.								MSU
Mollusca	Gastropoda	12-16 MI Manistee Co. Manistee R.								MSU
Mollusca	Gastropoda	12-17 MI Iosco Co. AuSable R.								MSU
Mollusca	Gastropoda	12-18 MI Kent Co. Grand R.								MSU
Mollusca	Gastropoda	12-19 MI Manistee Co. Manistee R.								MSU
Mollusca	Gastropoda	12-20 MI Kent Co. Grand R.								MSU
Mollusca	Gastropoda	12-21 MI Menominee Co Menominee R								MSU
Porifera	Spongillidae	12-22 MI Newago Co. Muskegon R.								MSU
Nematoda		12-23 MI Kalamazoo Co. Kalamazoo R.								MSU
										MSU
										MSU
										MSU

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