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Benthic macroinvertebrates as indicators of water quality: The intersection of science and policy

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Summary

This review addresses the intersection of water quality policy and benthic macroinvertebrates. Specifically, we examine the role that stream macroinvertebrates have played or could play in informing water quality decisions given the current policy framework, using this framework as the organizational structure for the review. Macroinvertebrates, as biological indicators of stream water quality, can be utilized to identify impaired waters, determine aquatic life stressors, set pollutant load reductions, and indicate improvement. We present both current approaches as well as innovative approaches to identify macroinvertebrates and aquatic life stressors. We also discuss an example of the environmental management approach, specifically, how macroinvertebrates can be used to indicate the relative success of stream restoration. For policymakers, this review serves to illuminate opportunities and limitations of using benthic macroinvertebrates as indicators of water quality. For entomologists, this review highlights policy-relevant research questions that would further aid the classification of impaired waters, the identification of stressors, or the management of stream ecosystems.

Keywords

Biocriteria; biological criteria; Clean Water Act; biological monitoring; bioassessment; stressor identification; stream restoration; benthic macroinvertebrates

1. Introduction

Clean fresh water is a basic human need as well as an important natural resource. Protecting or improving water quality is a great concern to governments around the world. Yet, in the United States (U.S.), recent surveys determined that 44% of sampled

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stream miles were polluted (United States Environmental Protection Agency, USEPA, 2009), and that 42% of U.S. wadeable streams and rivers were in poor condition while only 25% were in fair condition when compared to ecoregion-specific reference conditions (Paulsen et al., 2008). This suggests that a significant pollution problem remains regardless of the success stories of improved waterbodies. A number of notable water quality improvements occurred by regulating point source inputs, which resulted in technological improvements to wastewater treatment and the establishment of the National Pollutant Discharge Elimination System (NPDES) permits. But, as demonstrated by the recent studies of stream and river health in the U.S. (USEPA, 2009; Paulsen et al., 2008), water quality continues to be degraded by nonpoint pollutant sources. Thus, developing and refining approaches to identify and treat degraded waterbodies needs to continue.

There are several ways to assess water quality in lotic (flowing waters such as streams) and lentic (still waters such as lakes) waterbodies; the most common methods focus on physical and chemical (i.e., physicochemical) properties, such as the level of dissolved oxygen, mercury, and water clarity (priority pollutants listed in CWA section 307(a) in addition to those set by the state). Physicochemical parameters, which provide snapshots of the condition of a waterbody, do not provide the integrative measure of overall health of a stream and can, at times, inadequately identify impaired waters (United States Environmental Protection Agency, USEPA, 2005). Instead, biological measures provide an integrated, comprehensive assessment of the health of a waterbody over time (Karr, 1999). These biological indicators, also called biocriteria, use measures of the biological community including lower trophic level organisms, such as algae or benthic macroinvertebrates, as well as upper trophic level species, such as fish.

In this review, we present the intersection of benthic macroinvertebrates and ambient water quality policy in stream ecosystems. We introduce the water quality policy framework used to list impaired waters and to reduce pollutant inputs. We then describe how macroinvertebrates are currently used or could be used to list impaired waters, identify causes of impairment, set goals for reducing impairment, and indicate improvement in water quality. Specifically, we discuss the role of benthic macroinvertebrate data and monitoring in developing biocriteria and subsequently identifying the cause of water quality impairments in streams. We present both commonly used methods and more innovative approaches. We focus on streams because the use of macroinvertebrates as biological indicators is better established in lotic systems. We conclude with a list of recommendations for both scientists and policymakers suggesting productive future research directions that will facilitate and strengthen collaboration between these fields to improve the use of macroinvertebrates for water quality assessment.

2. Policy framework: Clean Water Act and biocriteria

U.S. waterbodies are regulated by both federal and state[1] governments. The Clean Water Act (CWA) is the federal policy that protects ambient water quality, but states

[1] Note: The use of state refers more broadly to individual states, tribes, and U.S. territories.

are given jurisdiction to monitor waterbodies, to list impaired waters, and to oversee the implementation of pollutant reduction strategies. The U.S. Environmental Protection Agency (USEPA) ultimately approves each state's criteria, list of impaired waters, and any decisions to delist impaired waterbodies. Thus, states have the authority to choose how they manage their waters and they have not adopted one uniform approach. There is, however, a general framework for managing ambient waters that indicates state versus federal authority; we detail this framework below (Figure 1).

The goal of the CWA (United States Code title 33, sections 1251-1387) is to "restore and maintain the chemical, physical, and biological integrity of the Nation's waters (section 1251)." Thus, the CWA requires that impaired waterbodies be identified and subsequently improved (Figure 1). Impaired waterbodies are identified using water quality standards. Water quality standards have four components: a narrative designated use, qualitative or quantitative criteria, the antidegredation clause, and general policies (40 Code of Federal Regulation (CFR) sections 131.10- 131.13). The narrative designated use describes the water quality goal. The CWA specifies an interim goal that all waters should meet: the waters should be fishable and swimmable. States have the authority to (and often do) set additional designated use classifications such as public water supply, primary contact recreation, and warm water fisheries. Because the designated use cannot be directly assessed, criteria are used as a scientific surrogate for the designated use. Criteria can be both physicochemical (e.g., total nitrogen, mercury, or totals suspended solids) or biological (e.g., chlorophyll *a* or index of biological integrity) metrics. When we use the term criteria, in this paper, we are referring more generally to any physical, chemical, and/or biological measures of stream health; when we use the term biocriteria, we are referring solely to biological measures. Though numeric criteria minimize difficulty in detecting and listing impaired waters, criteria can also be narrative descriptions of the conditions desirable for the use, such as "…a wide variety of macroinvertebrate taxa should be normally present and all functional groups should be well represented…" (State of Connecticut Department of Environmental Protection, 2002).

The third component of water quality standards is the antidegredation clause, which requires that a waterbody cannot be degraded below the point where it does not meet its current or existing uses (i.e. existed on November 1975 onward) (40 CFR 131.12). The general policies are directions describing the implementation of the standard, such as variance, low-flow policies, and mixing zones (40 CFR 131.13). The USEPA oversees and approves the standards set by the states (40 CFR sections 131.4 and 131.5). If a state does not set criteria or does not set criteria that the USEPA agrees are appropriately protective, the USEPA can assert jurisdiction and impose criteria (40 CFR sections 131.31-131.38).

The use of criteria as proxies for the designated use has emphasized the need to better demonstrate the linkage between the designated use and the criteria (National Research Council (NRC), 2001; Reckhow et al., 2005). Thus, the use of biocritieria as an additional indicator of waterbody health for designated uses focused on aquatic life use has gained increased attention (United States Environmental Protection Agency, USEPA, 1998) because these indicators provide an integrated assessment of the waterbody's health and have the potential to identify system degradation before it is detected by physicochemical criteria.

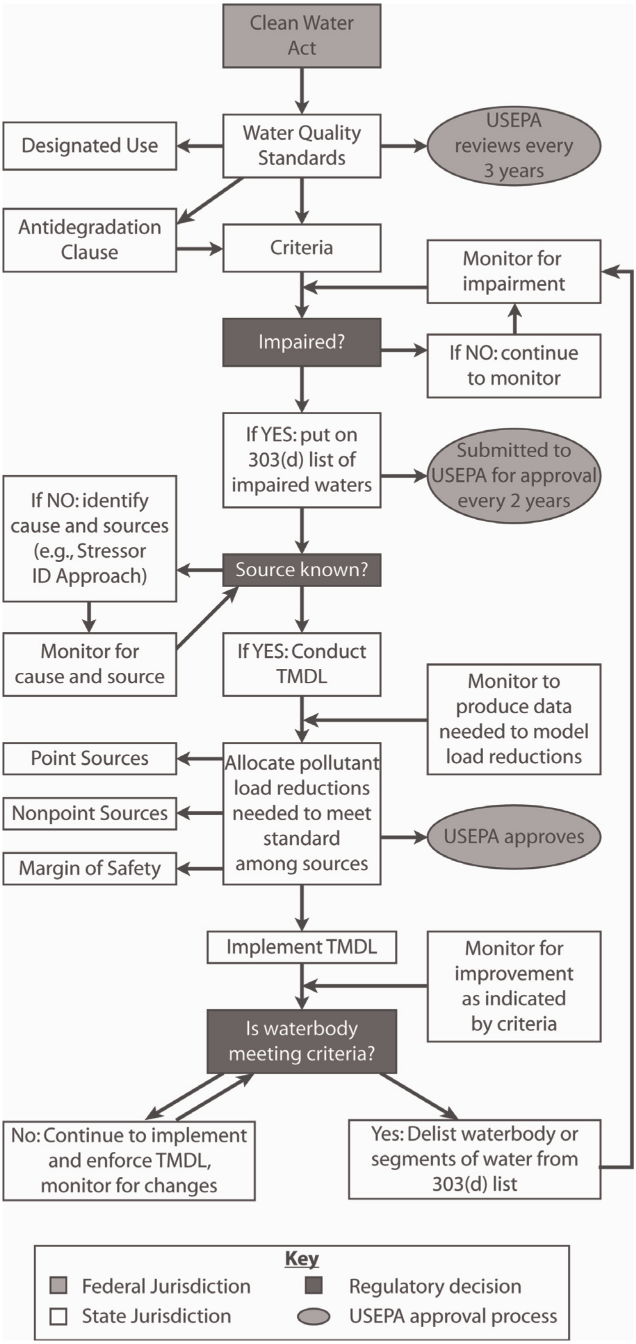

Figure 1. Policy framework describing the process to implement the Clean Water Act. Specifically the process involves detecting impairment, identifying causes, developing goals to reduce impairment, and improving the water quality to meet the criteria; the diagram indicates whether state or federal government has jurisdiction over the action. The diagram was created by the authors using information from U.S. Code title 33, sections 1251-1387 and USEPA (1994).

Once established, criteria are used to decide whether or not to list a waterbody as impaired (see section 4). When a state determines that a waterbody is not impaired, it continues to monitor regularly and check impairment status. When a waterbody is classified as impaired, then the state lists it on the 303(d) list of impaired waterbodies. This list is submitted by the state to the USEPA for approval every two years.

If a waterbody is listed as impaired, then action must be undertaken to improve the water quality such that it attains the designated use, as measured by the criteria. The state determines whether or not the source of the problem is known before determining the pollutant reductions necessary to meet the criteria. If there is a biological impairment and the problem is unknown, then the state conducts an analysis, such as the Stressor Identification (SI) process, to identify the causes (see section 5). Current data may be sufficient or additional monitoring data might be needed to identify the causes and sources. Once the causal pollutants are known, the state conducts an analysis to establish the total maximum daily load (TMDL) (see section 6). The TMDL sets a maximum pollutant load that still supports the designated uses and is approved by the USEPA. TMDL implementation involves actions such as improving pollutant reduction technologies at point sources or encouraging the establishment of various best management practices (BMPs) designed to reduce nonpoint source pollutant loading. During and after TMDL implementation, the state continues to monitor the waterbody for improvement. Waterbodies that do not meet their criteria remain on the 303(d) list, and the TMDL implementation and enforcement continue along with monitoring. A waterbody or a segment of the waterbody that has been assessed to meet the criteria is delisted, but continues to be monitored as part of the standard waterbody assessments.

3. Macroinvertebrates and stream ecosystem assessments

An important application of our ecological knowledge of stream macroinvertebrate communities is the bioassessments of stream ecosystem health. Bioassessment protocols are based on the premise that biotic communities respond to changes in habitat and water quality resulting from anthropogenic disturbance and that such community responses are integrated indicators of the state of the biotic and abiotic variables representing stream health (Bonada et al., 2006; Karr, 1999; Karr and Chu, 1999; Rosenberg and Resh, 1993). Barbour et al. (1999, pg 1-1) define bioassessments as "an evaluation of the condition of a waterbody using biological surveys and other direct measurements of the resident biota in surface water." Biological monitoring, as defined by Karr and Chu (1999, pg 2) includes "measuring and evaluating the condition of a living system, or biota" and is a process occurring over time designed to "detect changes in living systems, specifically, changes caused by humans apart from changes that occur naturally" in order to identify ecological risks to humans. Thus, bioassessments are individual evaluations of stream ecosystems and important components of long-term biomonitoring projects. While fish, algal, and macroinvertebrate assemblages each have particular advantages in bioassessments (Barbour et al., 1999), stream macroinvertebrates are most commonly used due to the simple equipment needed to sample them and the comparative ease of the sample processing. Additionally, because macroinvertebrates are tipically less mobile than fish, macroinvertebrates provide a more localized assessment of their response to stream conditions (see Barbour et al., 1999 for list of advantages and disadvantages for each taxa). Freshwater benthic macroinvertebrates include representatives of many insect orders, as well as crustaceans, gastropods, bivalves and oligochaetes (Allan, 1995; Merritt et al., 2008; Thorp and Covich, 2001), and they contribute to many important ecological functions, such as decomposition, nutrient cycling, as well as serve an important role in aquatic food webs as both consumers and prey (Covich et al., 1999; Moore, 2006; Vanni, 2002; Wallace and Webster, 1996). However, insects are often the dominant group of benthic macroinvertebrates in both absolute numbers and species diversity, which is not surprising given that the juvenile stages of many terrestrial insects are typically aquatic (Merritt et al., 2008).

The structure of macroinvertebrate communities depends on abiotic and biotic factors that vary across spatial scales from regional to habitat-specific and is discussed in detail by Lamoureaux et al. (2004), Malmquist (2002), Poff and Ward (1990), Vannote et al. (1980), and Vinson and Hawkins (1998). The natural features of stream and terrestrial habitats can affect macroinvertebrate assemblage structure. These features include: 1) the quality and quantity of food resources, 2) habitat quality such as the physical structure of the stream bed, 3) flow regime such as the frequency and intensity of storm-flow disturbance, 4) water quality, 5) biotic interactions, and 6) the condition of the riparian zone (see summary by Karr (1991), Mackay (1992), Sweeney (1993), Townsend et al. (1997), and Wallace et al. (1997)). Agricultural and urban land-uses greatly alter both the physical and the chemical aspects of macroinvertebrate habitat, impacting the structure of macroinvertebrate communities (Allan, 2004; Moore and Palmer, 2005; Paul and Meyer, 2001; Walsh et al., 2005b). Figure 2 presents an illustrative example of how macroinvertebrate communities can respond to land-use change through a chain of indirect effects that lead to changes to the macroinvertebrate assemblage in both taxa richness and relative abundance (Norris and Georges, 1993). These relationships between macroinvertebrate communities and stream ecosystem conditions make community structure a good indicator of overall stream health (Karr, 1999).

Bioassessments assume that macroinvertebrate community composition changes along a gradient of stream habitat and water quality (Resh et al., 1995) and that judgments of stream health can be made in relation to reference conditions (Barbour and Gerritsen, 2006). The "reference condition", as defined by Stoddard et al. (2006), describes a point of reference against which to compare the current state of a site. Ideally, reference conditions represent the naturally occurring physicochemical and biological conditions present in the absence of significant human impact. Stoddard et al. (2006) define a range of reference conditions, such as minimally disturbed condition, historical condition, least disturbed condition, and best attainable condition. However, few stream ecosystems are free from some sort of human impacts, which makes defining reference conditions even more necessary when applying the approach to bioassessments (Walter and Merritts, 2008).

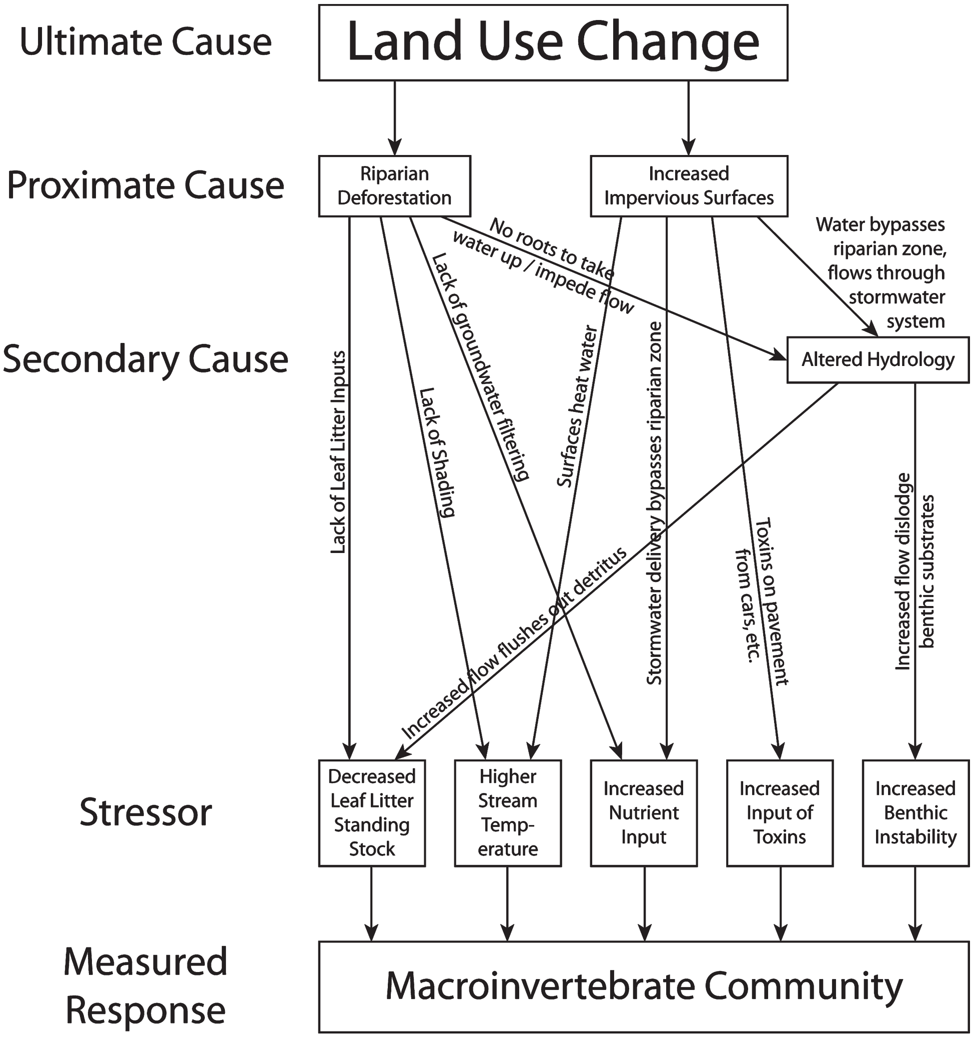

Figure 2. Illustrative schematic of the potential interactions between the causes, the stressors, and the response of the stream macroinvertebrate assemblage. This schematic is not intended to show all possible interactions and effects (see Karr, 1991). This diagram is intended to show the lack of direct links between any single stressor and even a few of the many potential causes. Though not presented here, a similar diagram could be developed for an agricultural system.

Bioassessments may utilize various indicators including single metrics, multimetric indices, or more complex multivariate predictive indices (Bonada et al., 2006; Karr, 1999; Karr and Chu, 1999; Rosenberg and Resh, 1993). Many different individual measures of macroinvertebrate communities are used in bioassessments and many are based on population and community ecological theory. Abundance and richness of assemblages or communities are simple measures and are often used in assessments; species-poor systems are generally assumed to have degraded water quality (Norris and Georges, 1993). Certain taxa, such as stoneflies (Plecoptera), are known to be more sensitive to pollutants or other stressors (DeWalt et al., 2005) and their presence is often considered an indicator of a healthy stream. Groupings of sensitive taxa such as the presence of EPT, which measures the proportion of individuals in the orders Ephemeroptera (mayflies), Plecoptera (stoneflies), and Trichoptera (caddisflies) are also used as an indicator of a healthy stream. Metrics to measure stream health can also assess the relative abundance of macroinvertebrates in groups such as feeding mode (i.e., functional feeding groups) or habitat niche (Barbour et al., 1999; Bonada et al., 2006; Karr, 1999; Rosenberg and Resh, 1993). Barbour et al. (1999) provides an extensive list of metrics and citations documenting their development, and for a more in depth discussion of stream macroinvertebrate bioassessment indicators, we recommend the following citations: Bonada et al. (2006), Karr, 1999 (1999), and Rosenburg and Resh (1993).

The choice of sampling and analysis methods impacts the conclusions drawn about impairment (Downes et al., 2002). Sampling design should maximize variation in biological indicators due to site-specific conditions; it should maximize the "signal" (i.e., response) relative to the "noise" (i.e. natural regional and temporal variation) and minimize the error variation associated with the sampling process (Barbour and Gerritsen, 2006). Numerous questions related to the sampling procedures including

1) whether to use qualitative or quantitative sampling methods,
2) what habitat(s) to sample,
3) how much and at what scale should replication be done, and
4) when to sample,

all have important implications for the spatial and temporal extent of the sampling. The spatial and temporal aspects of stream sampling designs influence the relative strength of "signals" from anthropogenic sources versus "noise" from natural sources of biological variation in the biota (Fend and Carter, 1995; Wiley et al., 1997). Partitioning out these two types of variance is important for determining the true effect to communities from anthropogenic sources (e.g., land-use change) versus natural changes in the macroinvertebrate assemblage. Probabilistic sampling assesses stream and river networks by first organizing reaches into groups by using characteristics such as stream size, and then, for each sampling event, selects a random sample of sites within each group. This approach is designed to reduce the bias in estimating the ecological condition of water resources (i.e., the anthropogenic impacts to waterbodies) within a larger region, based on a limited number of waterbodies sampled (Barbour and Gerritsen, 2006). In contrast, stream ecosystem sampling plans for long-term and large-scale impacts, such as climate change, may require different sampling methods in order to quantify unique responses despite ecosystem variability (Hauer et al., 1997). In addition to the sampling design, questions related to sample processing procedures including:

1) What, if any subsampling is needed?,
2) What sorting procedure to use to remove specimens from sample debris?,

3) What taxonomic level should specimens be identified to?, and
4) Should all macroinvertebrates be included in the analysis?,

will also have important implications for the type of data obtained from the surveys (Carter and Resh, 2001).

It is not uncommon for states have both targeted and probabilistic sampling programs. The targeted or fixed-station sampling provides long-term data at a fixed location so that one can observe changes over time. The probabilistic sampling gained attention because it provides an unbiased measure of the stream condition, improving the data provided to Congress through the National Water Quality inventory (Clean Water Act, section 305(b)). The important thing is that the sampling method chosen should be appropriate to conduct the bioassessments and then the data should be applied to identify impairment using the biocriteria.

4. Intersection of science and policy

4.1. Identifying impaired waters

While biocriteria are used to classify whether or not a waterbody's designated uses are impaired (Figure 1), deciding how to set biocriteria is difficult because it involves techniques from both bioassessment and ecological risk assessment (Suter, 2001). Bioassessments define ecological status. Risk assessment links stressors to attributes (both environmental and socio-economic) valued by society, and it quantifies or describes the outcome of each attribute given a range of criteria decisions. A policymaker can compare the risks and benefits and set a criterion threshold level that manages for these sometimes competing factors (Kenney et al., 2009; Suter, 2001). This process is how numeric criteria, either implicitly or explicitly, are set. These criteria levels are used to list which waters are impaired and seeks an appropriate balance between identifying and improving impaired waterbodies without wastefully spending resources to further research and improve misclassified waters.

4.2. Current approaches to set criteria: bioassessment

Quantification of a stream's ecological condition draws upon a variety of numeric metrics, described earlier. These indicators are derived from macroinvertebrate assemblage data, which are selected to indicate the degree of attainment of the aquatic life designated uses. There are a number of approaches used to aid in setting biocriteria. A common approach uses EPT. While not all species of EPT taxa are sensitive to pollution, the abundance of taxa in these orders gives a reasonable indication of stream health. In comparison, biotic or tolerance indices, a more complex method for determining the ecological condition of a stream, is a well-established approach that uses a weighted average of the abundances of taxa at a site multiplied by the predetermined taxon-specific tolerance values for particular stressors (Bonada et al., 2006). Tolerance values are a measure of pollutant sensitivity developed regionally by aquatic biologists and are assigned to an individual taxon based on the location of that taxon's peak abundance in streams along a stressor gradient. Such individual metrics, which often assume a simple linear response to degradation, can be standardized and aggregated to create a multimetric index value. A multimetric index derives a single score that aggregates multiple single metrics or biotic indicators that each change in a linear fashion along a stressor gradient (Karr, 1999). Impairment of a site is judged relative to the distribution of multimetric scores for undisturbed reference sites.

Rapid Bioassessment Protocols (RBPs) are widely used in conjunction with multimetric analyses (Barbour et al., 1999) and emphasize quick and efficient field sampling protocols and streamlined laboratory procedures to provide the needed inputs for multimetric indices. This approach is commonly used because it provides resource managers with understandable results at a minimum cost (Bonada et al., 2006). Although RBPs are consistently able to distinguish benthic assemblages from different geographic regions and to detect severe pollutant impacts, the RBP evidence is not sensitive enough to detect low-level or incipient impacts of nonpoint source pollution (Taylor, 1997). Identifying the level of taxonomic precision as well as the sampling design characteristics (e.g. size and number of replicate samples) necessary to accurately assess impairment provides a way to balance between maximizing data while minimizing costs (Jones, 2008). Sensitivity is often increased with species-level data, but family- or order-level data is appropriate to detect severe impacts (Taylor 1997, Jones 2008).

Multimetric indices are robust indicators that summarize a range of environmental responses and are usually understood by resource managers and the public (Karr and Chu, 1999). Multimetric methods, however, are less capable of distinguishing between impacted and reference sites than multivariate assemblage methods (Reynoldson et al., 1997). Instead of summarizing macroinvertebrate assemblage structure in a single index developed from individual metrics, multivariate approaches consider all the biotic conditions of a site together while summarizing the relationships between taxon abundances (i.e. presence and/or absence), environmental variables, and reference conditions. The multivariate method known as RIVPACS uses probabilities of detection based on reference conditions to develop a list of expected taxa which is then compared to the observed taxa in order to make assessments about the stream (Clarke et al., 2003). This method, and other approaches such as those developed in Australia (AUSRIVAS) and Canada (BEAST) are complicated analytical tools that for brevity, will not discussed here (see Hawkins et al., 2000; Reynoldson et al., 1997 for a comparison).

Though most states use bioassessments and have adopted narrative biological criteria, the majority of them have not adopted numeric biocriteria even though these criteria can predict benchmarks of aquatic life designated uses (United States Environmental Protection Agency, USEPA, 1991; United States Environmental Protection Agency, USEPA, 2002). It is not uncommon for there to be tiered aquatic uses to further define the aquatic life condition expected along a biological gradient. Of the states that have adopted biocriteria, Ohio and Maine have two of the more established, well-regarded programs. Ohio uses a multimetric biological index that is based on reference conditions, an approach that is consistent with the USEPA guidance (United States Environmental Protection Agency, USEPA, 1996). Maine uses a multivariate linear discriminant model (http://www.maine.gov/sos/cec/rules/06/096/096c579.doc) that quantifies the likelihood that a sample falls into one of the four tiered aquatic life classes (Davies and Jackson, 2006). For comprehensive summaries of each state's status in developing biocriteria, we recommend USEPA (1991), USEPA (2002), and Shelton et al. (2004).

4.3. Biomonitoring to detect impairments

Once an impaired waterbody is identified, regulatory steps must be taken to restore its integrity. The most desirable indicators for bioassessment are those that change at a point where the ecological structure or function of the stream changes significantly due to a stressor. Although the observation by Klein (1979) of a sharp change in macroinvertebrate community composition at or above 10% watershed cover by impervious surfaces is often cited as an example of such a threshold response to urbanization, subsequent macroinvertebrate surveys by Morse et al. (2003), Ourso and Frenzel (2003), Bonada et al. (2006), Moore and Palmer (2005), and Gresens et al. (2007) have detected responses at even lower levels of imperviousness, implying a linear response. To detect such changes, a sufficient sampling distribution along the various levels of impact is important to detect the environmental responses despite the presence of natural variability (Gresens et al., 2007). Nevertheless, nonlinear threshold responses of biological indicators (i.e. those responses where a unit change in one variable has less or more than a unit change in the other variable) are useful for defining a criterion level when there is a strong single threshold response; it is more difficult when the multimetric indices exhibit multiple change points in response to several stressors that vary in intensity (King and Richardson, 2003; Stevenson et al., 2008).

The taxonomic precision influences the conclusions drawn about stream health. For example, in Queensland, Australia the need for species-level data was evident when family-level identifications biased the results toward a higher level of stream health then was warranted given the species-level tolerance data for Chironomidae (non-biting midges) and Plecoptera (stoneflies) (Haase and Nolte, 2008). Despite the benefits of greater taxonomic resolution, species-level identification of juvenile insects is difficult, in part because species keys largely apply to adult life stages and these stream health assessment methods are not suited for adult stages (DeWalt et al., 1999). Therefore, the continued development of new and/or nontraditional taxonomic identification methods is vital for increasing the quality of bioassessments. Two such innovative approaches, not commonly used, include chironomid pupal exuviae (see Box A) and molecular methods (see Box B). These methods may decrease either the time or cost of sampling macroinvertebrates and therefore facilitate the use of macroinvertebrate data in bioassessments. Continued development of these nontraditional methods provides new research opportunities and challenges for insect taxonomists and entomologists (see section 7).

Box A. Innovative techniques for identifying macroinvertebrates: chironomid pupal exuviae

One current approach to improve the identification of individuals in macroinvertebrate sampling is the examination of chironomid pupal exuviae (Calle-Martínez and Casas, 2006; Raunio et al., 2007; Wilson and Bright, 1973; Wilson and McGill, 1977). Chironomid assemblages can have high species diversity, yet they are often identified only to family level in stream bioassessments (Lamouroux et al., 2004). The larval Chironomidae assemblage alone can indicate impacts associated with pollutants from agricultural and urban land use (Lenat and Crawford, 1994; Paul and Meyer, 2001); they are particularly useful for identifying moderately impacted streams because chironomid density and richness remain high even after sensitive EPT taxa decline (Coffman and de la Rosa, 1998; Maasri et al., 2008). Nevertheless, larval Chironomidae approaches are less utilized than EPT and other such approaches because: 1) the larvae demonstrate small morphological variability compared to "EPT" taxa and 2) the process of slide-mounting mature larvae head capsules to make genus- or species–level identifications is laborious.

An alternative method for collecting chironomid genus- or species-level data is to collect the pupal exuviae. The cast pupal exoskeleton is collected by skimming the water surface with a shallow pan or a drift net after the emergence of the adult (Ferrington et al., 1991; Wilson and Bright, 1973). The translucent exuviae can provide genus- or species–level identification using a stereomicroscope, eliminating the need for slide-mounting (Coffman, 1995). Keys to both European (Langton and Visser, 2003) and North American (Ferrington et al., 2008) chironomid pupal exuviae are available. Additionally, the chronomid pupal exuviae approach provides an integrated sample across many stream habitats and can be more cost-effective than benthic samples (Ferrington et al., 1991; Raunio and Anttila-Huhtinen, 2008). Thus, this approach is particularly well-suited for sampling of large, non-wadeable rivers (Franquet, 1999).

5. Identifying causes and setting goals for reducing impairment

Identifying the cause of the impairment is essential to improve the condition of biologically impaired waterbodies. Sometimes, these stressors can be easily identified or identified using bioassessment approaches previously discussed. For example, Cormier et al. (2002) noted, for two impaired river reaches, that defining the component indicators of multimetric indices is useful for stressor identification. Component metrics such as percent tolerant taxa are ambiguous because changes can be due either to decreases in sensitive species or increases in particular tolerant taxa (Cormier et al., 2002). As a result, the lack of understanding of life history specializations and ecological requirements of benthic insects provided by taxonomic assemblage data limits conclusions drawn about the cause of impairment (Cormier et al., 2002; Jones, 2008). This problem can be remedied, in part, by expanded knowledge of the ecological

Box B. Innovative techniques for identifying macroinvertebrates: molecular analysis approaches

Molecular methods are now being developed to improve benthic macroinvertebrate identification. Such approaches quantify variation in DNA nucleotide sequences (Hebert et al., 2003). Following DNA extraction from a specimen, two different molecular methods are used to discriminate aquatic insect species: 1) polymerase chain reaction (PCR) followed by analysis of restriction fragment length polymorphisms (RFLP) (Carew et al., 2007; Sharley et al., 2004) or 2) sequencing of DNA from the cytochrome oxidase 1 (COI) gene, referred to as DNA barcoding (Ball et al., 2005; Sinclair and Gresens, 2008; Zhou et al., 2007). These methods require extensive libraries wherein genetic sequence data are associated with specimens identified to species by traditional morphological taxonomy. Such libraries are being constructed for order-level Ephemeroptera (Ball et al., 2005), Trichoptera (Zhou et al., 2007) and numerous genera of Chironomidae (Carew et al., 2007; Ekrem et al., 2007; Sharley et al., 2004; Sinclair and Gresens, 2008).

Despite the existence of nearly universal primers, current DNA sequencing methods still require analyses of individual specimens because correctly aligning and interpreting gene sequence data obtained from a mix of species is not feasible. The reported cost for DNA gene sequencing is at least 5 to 10 US dollars per direction of DNA sequence per specimen (Ball et al., 2005). Whether this approach is cost-effective depends on how much time is needed to identify difficult specimens using an expert taxonomist (Ball et al., 2005; Carew et al., 2007). In addition, taxonomic groups, whose species are not well-defined by traditional systematic methods, are also not reliably identified by DNA barcoding (Alexander et al., 2009). Thus, the continued development of aquatic macroinvertebrate taxonomy mentioned previously is also important for DNA barcoding. Thus, increasing collaboration between researchers in molecular systematics and specialists in bioassesments is important to promote the development of efficient, effective identification tools. (Jones 2008).

tolerances of aquatic organisms related to specific abiotic stressors. For example, King and Richardson (2003) recently established stressor-response relations of wetland macroinvertebrate assemblages to a phosphorus gradient by combining invertebrate data sampled along a natural P gradient with a concurrent in situ P-enrichment experiment. They used ordination scores of invertebrate assemblages along the P gradient and metrics based on species-specific tolerance values to the local P gradient to determine benchmark conditions (King and Richardson, 2003). Similarly, Smith et al. (2007) successfully developed genus- and species-level macroinvertebrate tolerance values that produced separate biotic indices to distinguish responses to total phosphorus and nitrate levels in streams. Therefore, to maximize the sensitivity and diagnostic value of tolerance values, the approach should be refined in the following ways:

1) develop new sets of tolerance values specific to particular stressors, 2) define, when possible, tolerance values at lower taxonomic levels (i.e. species), and 3) tailor tolerance values to regional variation (Resh and Jackson, 1993; Yuan, 2007; Yuan and Norton, 2003).

Distinguishing the effects of multiple stressors using macroinvertebrate assemblage structure data is difficult because of the web of indirect effects and interactions between ultimate causes of ecosystem degradation and the proximate stressors of the assemblage (Figure 2) (Allan, 2004). For example, a decrease in the abundance of individuals belonging to the shredder functional feeding group would seem to indicate some type of impact resulting from decreased leaf litter input. However, impacts to shredders are likely to transfer to other trophic levels and functional feeding groups through trophic interactions (Wallace et al., 1997). Riparian deforestation, a cause of decreased litter inputs, may also cause altered temperature regimes, increased nutrient inputs, increased flashiness, decreased bed stability, and increased sedimentation, which also all may affect shredders and other functional feeding groups (Paul and Meyer, 2001; Sweeney, 1993). Interactions among stressors may have additive, synergistic, or antagonistic effects on stream macroinvertebrates (Darling and Cote, 2008; Townsend et al., 2008). Some stressors may interact with natural sources of mortality (e.g. predators) to increase the effect of the stressor on stream macroinvertebrates (Schulz and Dabrowski, 2001). But there are several methods of identifying stressors using macroinvertebrate responses to particular pollutants or stressors. One method, which is more commonly used in Europe, is the biological traits method (see Box C). Another promising method is toxicogenomics (see Box D). Both of these methods could greatly improve our ability to identify particular stressors. Stressors may also interact through time, and legacy effects may play a role in determining stream invertebrate community structure (Harding et al., 1998; Walter and Merritts, 2008).

Stressor identification (SI), also known as a "lines of evidence approach" (Downes et al., 2002), is a logical process of organizing and analyzing a wide array of biological, chemical, and physical data in order to make causal inferences about human impacts on ecosystems. The goal of SI is to minimize the uncertainty as to whether an observed impact was caused by a particular stressor or by confounding natural variation (i.e. inferential uncertainty). This is accomplished by eliminating unsupported causes from a list of possible anthropogenic and natural causes for a specific impact, and assessing the support from potential causes using multiple, independent sources of evidence of causation (Downes et al., 2002; United States Environmental Protection Agency, USEPA, 2000). The SI process is similar to the process that a medical doctor might use to determine the cause of an ailment given the patient's statement and any tests performed because the data interpretation relies heavily on expert judgment about the likelihood that impairment is caused by certain stressors. Formal application of the SI process to stream and river ecosystems is relatively recent (Clements et al., 2002; Cormier et al., 2002; Downes et al., 2002 and references therein). The USEPA encourages adoption of the SI process through its detailed guide,

Box C. Cutting-edge methods for identifying stressors: biological trait data

An innovative approach used to identify stressors is the biological trait method. The main concept behind the use of biological trait data is that dominant species traits closely relate to ecosystem function (Grime, 1998) and, particularly in aquatic environments, to environmental conditions experienced by the organisms (Lamouroux et al., 2004). The hope is that unique species traits are expressed in response to different environmental stressors (Lamouroux et al., 2004); if this is true then measuring the traits of a community can help identify particular stressors. Several authors have suggested that biological traits bioassessments and biomonitoring may better separate individual stressors than traditional community-based assessment methods (see for example Doledec and Statzner, 2008; Doledec et al., 1999). Charvet et al. (1998) found that the assemblages living in the more variable but less adverse habitat upstream of a wastewater treatment plant were smaller, shorter lived, and less mobile, with more descendents per reproductive cycle and more reproductive cycles per year than species living in more stable, but adverse habitats, downstream. These results demonstrated that changes in stream pollution can lead to changes in the functional traits of macroinvertebrate communities. There are several additional potential benefits of the biological traits method. For example, measuring traits can be more cost-effective than measuring species richness because describing the trait composition of a macroinvertebrate community requires fewer samples than determining species richness (Bady et al., 2005). Also, an accurate description of the abundance of biological traits requires less taxonomic expertise because a researcher can use species, genera, or family data (Bonada et al., 2006; Doledec et al., 2000; Gayraud et al., 2003; Lamouroux et al., 2004).

There have also been promising studies suggesting that the effects of pollutants may be determined using physiological responses of organisms, such as changes in respiration rates (Coler et al., 1999). For example, in situ bioassays transplant cages of organisms into a site for 24 hours in order to measure responses (such as rates of energy consumption) under different levels of pollutants (Damasio et al., 2008).

Although these biological traits methods show promise, they need additional development and testing in order to be broadly applicable. As a result, there remain a number of questions that need additional research. These include:

1) Is characterizing traits for late instars an appropriate way to represent organism-environment relationships (Poff et al., 2006)?
2) What is the importance of assessing biological traits together or individually (Gayraud et al., 2003)?
3) How do we select the traits when the measurements need to be fairly convenient and also relate to the underlying relationship between organisms and their environment (Poff et al., 2006)?

In order for biological traits to truly improve on current bioassessment methods, research must demonstrate clear links between specific traits and the health or condition of a waterbody. For example, stable isotope ratios, such as δ^{15N} values, which vary depending on the environment an organism is exposed to, could be an indicator of pollutant loading. ^{15}N is not a pollutant, but waterbodies receiving human and animal waste tends to have a higher δ^{15N} signature than areas not affected by human discharges (Saito et al., 2008). Therefore, increases in δ^{15N} values in crayfish, snails, and periphyton have been used to identify human and animal waste contamination around urban areas (Saito et al., 2008). Such studies demonstrate how trait-based methods can be used to help identify specific stressors.

Box D. Cutting-edge methods for identifying stressors: toxicogenomics

Another new approach in biomonitoring is the field of toxicogenomics. Toxicogenomics examines the toxicological responses of organisms to pollutants at the gene level (Carvan III et al., 2008; Watanabe et al., 2007). In particular, the use of microarrays for measuring gene expression variation in populations is a promising tool for answering ecological questions such as the effect of anthropogenic stressors on native populations (Gibson, 2002). The premise is that different stressors (e.g., a chemical) are likely to elicit different responses within a cell depending on the metabolic pathway(s) (Watanabe et al., 2007). The current goal in toxicogenomics is to find stressor-specific changes in gene expression related to conditions in the field (Gibson, 2002). Laboratory studies have shown that exposure to pollutants can be linked to the expression of specific invertebrate genes. Gene responses in *Daphnia magna* (water fleas) have been observed in response to oxidative stress, heavy metals, and organophosphate pollution (Damasio et al., 2008; Watanabe et al., 2007). Both grass shrimp (Brown-Peterson et al., 2008) and blue crabs (Brown-Peterson et al., 2005) were found to have altered gene expression in response to hypoxia.

In order to be a useful method for stressor identification, gene expression must remain constant or at least change predictably through space and time along natural gradients as well as in response to anthropogenic alterations to the environment. Yet field tests of gene expression across a gradient of stressor intensity are generally lacking. Examples using stream macroinvertebrates successfully in toxicogenomic field studies are, to our knowledge, completely lacking. Nevertheless, field tests with other organisms show very promising results (Fernandes et al., 2002; George et al., 2004). For example, Hook et al. (2008) found that measurements of gene expression were able to identify individual chemical stressors in rainbow trout when exposed to a mixture of chemical toxins. This suggests that examining gene expression may be a good way to identify individual stressors in environments with multiple anthropogenic impacts that otherwise have confounding effects on community composition and structure. This area of research has great potential for collaborative research between entomologists, geneticists, toxicologists, and stream ecologists.

While the methods for successful identification of environmental hazards using in situ biassays of macroinvertebrates have improved (Damasio et al., 2008), using gene expression does have some potential drawbacks that may limit the scope of its use for identifying stressors. Implementation may be difficult because the equipment, techniques, and personnel needed to perform these analyses are expensive (Hofmann and Place, 2007). In addition, gene sequence information for non-model organisms is rarely available and needs to be developed prior to field surveys (Hofmann and Place, 2007). However, the current requirement that microarrays be species-specific could be bypassed if sequences can be identified which are common across species and that differ in expression (Kassahn, 2008). Confounding and unrelated factors in the field may also affect the responses of organisms to pollutants (Damasio et al., 2008). Individual organism responses are affected by factors such as nutritional status, genetic differences, seasonal cycles, and life stage (Carvan III et al., 2008). Gene expression may also differ between tissues within an organism (Venier et al., 2006) or between organisms in different geographic areas (Lilja et al., 2008).

Although the field of toxicogenomics needs to address these limitations, the application of toxicogenomic methods, such as microarrays, to biomonitoring holds a great deal of promise for helping to identify stressors to benthic macroinvertebrate assemblages (Robbens et al., 2007). Specifically, the combination of more traditional measures of stream health, such as water quality variables or diversity indices, with laboratory or field bioassays could be particularly powerful and is worthy of further study (Damasio et al., 2008).

"Causal Analysis/Diagnosis Decision Information System" (CADDIS) (http://cfpub.epa.gov/caddis/index.cfm); however, the general approach is widely applicable to many areas of ecological and environmental analysis that are not amenable to experimental determination of causal factors. The three major steps in stressor identification are outlined below.

The first step is to define the negative effects of concern and their extent in space and time. Multiple correlated impacts should be analyzed individually to distinguish different causes and compare their relative importance. A list of all possible anthropogenic and natural causes is used to construct a conceptual model of possible pathways of causation, which includes direct effects, indirect effects, and confounding factors (United States Environmental Protection Agency, USEPA, 2000). The conceptual model incorporates both site-specific field data and information from a thorough literature review of relevant studies (Downes et al., 2002).

The second step takes an epidemiological approach; it uses the available data to eliminate as many candidate causes as possible (Downes et al., 2002). The types of causal indicators are 1) the strength of association between putative causes in time and space across a gradient of biological response, 2) the strength of the response to a measured stressor, 3) biological plausibility or the likelihood that the proposed mechanism can cause the stressor, and 4) specificity or the uniqueness of the symptom to the stressor (Groenendijk et al., 1998). The causal indicators are then used to develop a model which determines if data collected from the impacted site indicates impairment.

Lastly, causal indicators are used to build a qualitative ranking of the strength of evidence in support of each potential cause. Ideally, only a few hypothetical causes are left following the first two steps, and this last step will distinguish the most likely causal stressors. However, rather than one candidate cause, several possible causes might remain and causal inference cannot be made if none of the remaining causes receive strong support. If this is the case, additional data are gathered or collected to either support or eliminate these possible causes. The process is repeated with these new data until stressors have been conclusively identified.

6. Improving impaired waters: Total Maximum Daily Load (TMDL) designation

If a waterbody is on the 303(d) list of impaired waters, the state is legally obligated to reduce the pollutants of concern. The state develops a formal plan by determining the current system load inputs and then assigns a total maximum daily load (TMDL) that predicts the maximum loading that still achieves the criterion; the necessary load reduction is the difference between these two loads (Figure 1). This load reduction is then allocated to the point sources (PS) and nonpoint sources (NPS). Often, a margin of safety (MOS), which reserves a portion of the load allocation to account for uncertainty in the allocations, implementation, and future changes, is also allocated (equation 1).

$$TMDL = \sum PS + \sum NPS + MOS. \quad (1)$$

The TMDL report may additionally include descriptions about how the point sources will be required to meet and nonpoint sources will be encouraged to meet load reductions. The TMDL does not necessarily account for land use or other types of changes in the watershed that may impact pollutant levels. Both the uncertainty of these future inputs are usually accounted for using an appropriately conservative MOS. Even though a simple equation defines the TMDL, the loading values are based on models with sometimes substantial uncertainties.

The assessment and subsequent assignment of loads to sources is easiest when the stressor or stressors are known and can be incorporated into appropriate EPA-approved TMDL models. Identifying the cause of impairment using an approach such as SI (see section 5) is essential if the stressor is unknown. Once the pollutants are identified, additional monitoring data may be necessary to quantify model inputs. In some cases, current models may be inadequate to quantify the reductions necessary, and new methods may need to be developed to establish the TMDL.

An example of such a situation is a site impacted by multiple diffuse nonpoint sources. Maine recently tackled this problem by developing a novel TMDL that uses percent impervious cover as a proxy for a mixture of pollutants (Center for Watershed Protection (CWP), 2003; ENSR Corporation, 2005; Meidel and Maine Department of Environmental Protection, 2006). This approach provides some unique advantages. One, it takes advantage of the relationship between percent imperviousness and impact on aquatic life to define a target percent imperviousness as the TMDL. Two, it is not a pollutant-specific TMDL; it uses an impact standard that seeks to restore the aquatic life use instead of meeting the target level for a single pollutant (Courtemanch et al., 1989). Three, it provides Maine the ability to apply a suite of nonpoint source reduction options, such as BMPs, to improve waterbody condition. One such potential BMP is stream restoration (see Box E for more details).

For aquatic life, TMDL implementation is necessary to improve the condition of the waterbody as measured by the biocriteria. For point sources, the implementation of the TMDL is straightforward because the state can enforce mandatory pollutant reductions through effluent sampling. The implementation for nonpoint sources is more difficult because the allocation relies on voluntary measures to meet the reductions. Thus, in nonpoint source dominated systems, such as those with a significant amount of agriculture, there is no guarantee that the pollutant reductions will be sufficient to meet the TMDL. Without full TMDL implementation, the waterbody's conditions are predicted to deteriorate. Regardless of whether the TMDL is fully or partially met, reductions that notably change aquatic life as indicated by the biocriteria may take years. This may create difficulties in assessing the degree of success of various implementation strategies and determining the changes necessary to improve the likelihood of achieving the desired improvement.

7. Conclusion and future directions

This review presented the intersection of water quality policy and benthic macroinvertebrate science. Specifically, we highlighted the complex relationships between bioassessments using stream macroinvertebrates and their relevance for developing biocriteria, stressor identification, and TMDL implementation. We believe the intersection between these two fields provides opportunities and limitations for the policy and the science. We suggest research directions for scientists who want to help inform policy and policymakers who want to contribute to the scientific process.

7.1. Science contributions to policy

We believe that opportunities exist for macroinvertebrate ecologists to find new ways to apply community, population, and physiological information to bioassessments, biocriteria, and stressor identification. The following is a list of research opportunities and recommendations for entomologists and stream ecologists that would potentially improve the design and implementation of water quality policy.

Box E. Stream restoration: a management example

Stream restoration is a management option used to improve the health of a stream and has been more recently applied to mitigate pollutants to comply with consent decrees or TMDLs. Stream restoration can include any activity meant to alter the physical, chemical, biological, or aesthetic conditions of the stream to promote restoration goals (Bernhardt et al., 2005), but restoration activities often focus on modifications to the geomorphology and channel design (e.g. Rosgen and Silvey, 1996). The restoration design objectives, however, should be based on changes that will improve biotic or abiotic functioning (Reichert et al., 2007). Though setting meaningful objectives may sound intuitive, a nationwide survey conducted by the National River Restoration Study (NRRS) declared that 20% of stream projects had no listed goals (Bernhardt et al., 2005).

Biomonitoring of restored streams using macroinvertebrates is useful when restoration objectives include improving aquatic life, particularly restoring macroinvertebrate diversity and biomass. Macroinvertebrates are an integrative measure of stream health and, therefore, can be good indicators of the effectiveness of restoration. In addition, macroinvertebrates respond rapidly to restoration activities (Maloney et al., 2008; Stanley et al., 2002).

Several factors that should be considered in restoration design to promote the recovery of macroinvertebrate communities. One factor is the importance of habitat features. To improve the benthic community, common practice is to include design features that attempt to restore structural complexity and a diversity of stream habitats (Bernhardt et al., 2005; Hassett et al., 2005). For example, in-stream habitat complexity and adjacent riparian vegetation are two factors that determine the colonization potential of aquatic insects (Milner et al., 2008). Simply restoring habitat structure, however, does not always guarantee the restoration of community diversity and ecosystem function (Brooks et al., 2002; Lepori et al., 2005; Palmer, 2009; Palmer et al., 1997). Restoration of stream habitat may need to extend beyond the local habitat and consider the larger watershed (Palmer et al., 1997); stressors on the system are often due to features of the watershed. For instance in urban systems, watershed level impacts from impervious surfaces may indicate that restoration strategies need to focus on watershed scale stormwater drainage systems rather than reach level habitat manipulations (Walsh, 2004; Walsh et al., 2005a). Thus, determinations about restoring a stream to meet its criteria need to include an honest assessment of the ability to make the needed watershed level changes and/or the ability to engineer local changes to the stream that mitigate these watershed level impacts.

Another factor to consider when measuring macroinvertebrate communities to assess restoration success is the colonization potential of taxa. The ability to colonize a restored reach is dependant on the dispersal abilities of individuals, location of source populations, and the habitats traveled during dispersal (Bond and Lake, 2003; Lake et al., 2007; Young et al., 2005). Population colonization and recovery increase if source populations are nearby (Ahlroth et al., 2003; Fuchs and Statzner, 1990), but recolonization may occur on the order of years for more distant individuals that need to migrate to the restored reach (Milner et al., 2008). Assessing this recovery will require long-term macroinvertebrate community monitoring. In addition, long distance dispersal is most likely to occur during the adult stage. Thus, the features of terrestrial upland and riparian areas within the watershed can impact the movement of adult insects between streams (Smith et al., 2009). These large-scale watershed features may promote or prevent recolonization; therefore, these features must be considered in management or TMDL plans when streams struggle to meet their aquatic life use designation.

At the local scale, another factor which can be important for colonization and population persistence is species interactions. Milner et al. (2008), for example, demonstrated that large woody debris was important habitat for salmon, and that the scoured areas of salmon nests (redds) created disturbed patches which facilitated the persistence of early benthic macroinvertebrate colonizers. This suggests that aquatic insect diversity may be dependent on the colonization and survival of other species, such as fish, which serve as ecosystem engineers. Therefore, such interacting relationships need to be considered in the restoration design to maximize the success of both species.

Even though the scientific basis for restoration is still nascent and there is much to be learned from monitoring completed projects, long-term monitoring is still uncommon (Bernhardt et al., 2005). Better monitoring efforts not only will allow better tracking of restoration success, but also might help identify those stressors that cannot be mitigated through restoration efforts alone. Additionally, certain restoration activities might provide little or negative benefits (Palmer et al., 2005). A better understanding of what leads to restoration success or failure can allow limited resources to be spent on activities (either restoration and/or non-restoration approaches) that have the greatest likelihood of leading to a desirable outcome.

1. Can we develop improved methods for identifying larval aquatic insects including methods for extracting taxonomic data from assemblages (see Box A)? This involves both the continued development of methods (including keys) for identification of larval aquatic insects and the support of new methods such as the use of molecular methods as an alternative method for identifying aquatic insects (see Box B)
2. Make data more reliable and comparable across different regions to facilitate comparisons and to encourage data sharing between state agencies, universities, industries, and other research organizations. This will likely involve developing or maintaining support systems of certification (e.g. North American Benthological Society's certification program) that are accepted by states to increase reliability and comparability of datasets. Develop methods to standardize biological sampling protocols. Such data could be regularly uploaded into a central database, such as STORET (http://www.epa.gov/storet), to maximize access.

3. Are there applications of community ecology concepts (e.g. disturbance and successional dynamics, metacommunity and/or neutral theory) that can benefit the development of biocriteria? For example, how rapidly do community traits respond to environmental change? After a disturbance such as a change in land use or hydrology, how quickly does the community respond, and more specifically, how quickly do macroinvertebrate assemblage traits respond and demonstrate important changes in the assemblage?
4. Are there specific species traits that are linked to specific stream ecosystem functions? In other words, are certain traits better predictors of how well an ecosystem is functioning than others? Field trials of these relationships are very important. These trials will improve new species level methods using assemblage data for developing biocriteria (see Box C).
5. Which stressors can be detected best by in situ transplant experiments? What are the best physiological responses (such as respiration rates) to measure in organisms to detect stressors? Which organisms are best to use in transplant experiments?
6. How do organisms respond to different types of stressors and particularly to combinations of stressors? This is a question in which toxicogenomic studies may be particularly useful. Lab trials may be an important first step, but field trials of these methods will be necessary for this approach to be useful for water quality monitoring.
7. Is there a larger role for stable isotopes in biomonitoring efforts? Given, there are differences in the ^{15}N signature from human and animal-influence waste than communities without human inputs (see Box C), can stable isotopes be used more broadly to determine whether a community is stressed? How can we improve the process of identifying different types of stressors or combinations of stressors? Any of the new methods discussed in this review may provide future insights into stressor identification.

7.2. *Policy contributions to science*

Below we summarize a list of research opportunities and recommendations for policymakers that will improve the use of use biocriteria in the policy process.

1. Develop TMDL models that link specific stressors to aquatic life criteria. TMDL models are currently well developed to link physicochemical factors to specific stressors, but linking specific stressors to aquatic life biocriteria will improve TMDLs for biomonitoring.
2. Better understand the linkages between biocriteria and other water quality criteria such as nutrients, metals, and water clarity. An understanding of the interactions between these indicators may indicate non-linear combinations that negatively impact stream health before it is indicated by individual measures.
3. Continue to encourage all states to develop bioassessment databases that are useful for setting biocriteria or applicable to SI analysis. Many states do not have such databases, hindering the application of SI and the subsequent development of a TMDL. These databases should be shared to improve the information base

accessible to all the states. USEPA should regularly update this guidance to assist states in implementing such bioassessments.

4. Further explore the applicability and potential policy or implementation hurdles of using impact standards instead of performance standards to improve aquatic life uses (Courtemanch et al., 1989). Impact standards focus on the desired outcome instead of meeting a pollutant-specific target, making impact standards a particularly appealing alternative to improve the macroinvertebrate condition.
5. Improve current stressor identification protocols and develop novel approaches using macroinvertebrates to efficiently identify stressors. The identification of which pollutants are degrading a waterbody is essential to developing a TMDL and subsequently improving the waterbody. This step is key to better managing our waterbodies given land-use changes and increased impacts from nonpoint sources.
6. Develop and improve biocriteria methods for the non-flowing waters protected by the CWA. The majority of the research and application of biocriteria has focused on stream ecosystems, but the benefits of using biocriteria could also extend to lakes and wetlands.

The use of benthic macroinvertebrate indicators greatly enhances states' ability to identify and subsequently improve impaired waters, but there is still research needed. Collaboration between researchers and practitioners of entomology and environmental public policy could lead to novel research that is relevant to society and would further aid the classification of impaired waters, the identification of stressors, and the management of stream ecosystems.

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