A review of agricultural impacts on aquatic biotic integrity and recommended metrics to measure broad scale watershed improvements

Context of review

There is a general lack of information on aquatic communities in agricultural watersheds of the Midwestern United States, especially how biodiversity and community structure are affected locally by agricultural impacts such as degradation of instream habitat, reduced water quality, and loss of riparian vegetation. This review summarizes recent studies that have examined responses of aquatic communities to agricultural impacts in a specific attempt to identify biotic indices that were consistently responsive to these impacts, and thus can be most useful in measuring the effects of conservation practices implemented in agricultural watersheds. The literature review was limited to studies conducted at watershed- and statewide-scales within the Midwestern United States. Several statewide studies that surveyed a range of anthropogenic influences on nutrients and habitat conditions were also included (Miltner and Rankin 1998, Heatherly et al. 2007, Wang et al. 2007) because these studies were able to distinguish among watershed types (e.g., agricultural, urban, forested) and their associated impacts on biological communities.

Agricultural impacts on biotic integrity

Physical impacts of agricultural practices on streams and rivers include changes in riparian vegetation, channelization, altered hydrology, degraded instream habitats, and increased sediment and nutrient loads (Cooper 1993). The confounding effects of these multiple stresses have made it difficult to measure direct impacts of individual stresses on biotic integrity, community structure, and ecological function in Midwestern streams and rivers. In addition, the natural complexities of lotic ecosystem attributes (e.g., light limitation, spates, rapid nutrient cycling) increase the difficulty of compartmentalizing specific agricultural impacts and managing watersheds accordingly. Comparisons among studies that have examined the relationships between agricultural impacts and biotic integrity, however, have provided some consistent information that may be used to identify the relative importance of specific stresses to aquatic communities and the biological indices that are most sensitive to these stresses. In turn, these indices can be used to measure potential biodiversity improvements related to the implementation of conservation practices in agricultural watersheds.
**Riparian buffers.**- Most of the studies reviewed provide data that support the importance of intact, wooded riparian habitats to instream habitat quality, nutrient processing, and biotic integrity (Table 1). In southern Illinois streams, Stone et al. (2005) reported that even small amounts of riparian forest were associated with better instream habitat quality and biotic integrity. Statewide surveys in Illinois (Heatherly et al. 2007) and Wisconsin (Wang et al. 2007) also showed improvements in habitat quality and increased diversity and taxa richness of macroinvertebrate and fish assemblages with increasing forested land. In Iowa agricultural watersheds, Poole and Downing (2004) reported alarming declines of mussel species associated with the loss of riparian woodlands and reduced instream heterogeneous substrates. Collectively, these studies indicate that restoration of wooded riparian habitat is one of the primary conservation practices that may provide long-term protection of macroinvertebrates, fishes, mussels, and overall aquatic biodiversity from agricultural impacts.

**Instream habitat.**- From a stream management perspective, both the sources and instream processing of nutrients affect biotic integrity. Although adjacent land use and riparian buffers influence nutrient export to streams, instream habitat quality may also influence nutrient processing within the system (Miltner and Rankin 1998). Miltner and Rankin (1998) reported that habitat quality as measured by the Qualitative Habitat Evaluation Index (QHEI, Rankin 1995) explained 20-30% of variation in fish and macroinvertebrate biotic integrity scores across stream sizes and models. Similarly, macroinvertebrate biotic integrity scores were most strongly correlated with instream physical habitat quality in agricultural streams (Stone et al. 2005), in which USEPA’s Rapid Bioassessment (Barbour et al. 1999) was used to evaluate habitat. Strong links between habitat and biotic integrity suggest that instream physical habitat may be the most important factor limiting biotic integrity in agricultural streams. Thus, measurements of changes in habitat quality may provide early indications of improved ecosystem health in response to conservation practices.

**Nutrients.**- Recent research is revealing several trends with respect to changes in macroinvertebrate densities and assemblage structure with increasing nutrient concentrations and degraded habitat in freshwater systems. First, invertebrate densities and biomasses generally increased in streams that exhibit nutrient-enrichment (Miltner and Rankin 1998) and/or low forest cover (Stone et al. 2005). One exception was a study by Heatherly et al. (2007) in Illinois that found severely decreased densities of invertebrates in two severely impacted urban streams.
Second, higher invertebrate abundances and biomasses in degraded streams are typically a result of increased relative abundances of pollution tolerant taxa, such as non-tanypodinae dipterans and noninsects (Miltner and Rankin 1998), tubificid worms, fingernail clams (*Sphaerium*), and ostracods (Stone et al. 2005). Mayfly densities have also been reported to increase in impacted streams (Miltner and Rankin 1998, Stone et al. 2005), due to increased abundances of pollution tolerant genera such as *Caenis*, *Baetis*, and *Callibaetis* (Stone et al. 2005). Similarly, Cross et al. (2006) reported a significant increase in density, biomass, and secondary production of invertebrates in a paired-catchment experiment in which a headwater stream was experimentally enriched with nutrients over two years. For fish, Miltner and Rankin (1998) reported higher abundances in streams with intermediate nutrient levels, associated with decreased relative abundances of specialized feeders and increased proportions of tolerant or omnivorous species.

In many cases, limited interpretation has been made as to the direct effects of nutrients on biotic integrity because high nutrients generally occur where habitat quality is also degraded. However, several studies have provided evidence that supports local effects of nutrients on fish and macroinvertebrate biotic integrity (e.g., Miltner and Rankin 1998, Stone et al. 2005, Heatherly et al. 2007). In Ohio wadeable streams, Miltner and Rankin (1998) reported reduced fish index of biotic integrity (IBI) scores with increasing nutrient concentrations. They also found that the influence of total phosphorus (TP) on IBI scores appeared independent of habitat and other measured variables up to the median TP concentration. In a watershed survey conducted in northeastern Nebraska, Whilels et al. (2000) reported one of the lowest macroinvertebrate biotic integrity scores from a site that had the highest habitat quality, yet also the highest nutrient and suspended sediment concentrations. Similarly, Stone et al. (2005) demonstrated reduced scores for Hilsenhoff’s biotic index (HBI) for macroinvertebrate communities with increasing concentrations of orthophosphorus across 15 agriculturally impacted streams in southern Illinois. Heatherly et al. (2007) also suggest that phosphorus may be the critical nutrient affecting biotic integrity in Illinois agricultural watersheds where nitrogen concentrations are high enough to preclude algal nitrogen limitation. These studies suggest that the high nutrient concentrations commonly associated with agricultural watersheds throughout the Midwest may have direct effects on freshwater aquatic biodiversity. Whereas, management for nonpoint source nutrients in streams and rivers has often been conducted in relation to Gulf...
Hypoxia, new evidence suggests that high nutrient loadings may be impacting biotic integrity and ecosystem function at a more local level.

*Wetlands.* - Wetlands can also serve to buffer negative agricultural impacts on instream habitat quality, nutrient loadings, and aquatic biodiversity. However, in many agricultural areas of the Midwest more than 90% of the historic wetlands have been lost. As a result of wetland loss and increased use of tile-drainage, the rate at which water drains through these agricultural watersheds has dramatically increased which in turn increases instream erosion, degrades habitat quality, and increases nutrient inputs into aquatic ecosystems. Restoration of wetlands that retain tile-drained waters can effectively reduce nutrients (e.g., Kovacic et al. 2000), increase habitat and biodiversity, and has the potential to ameliorate altered hydrology.

**Summary of Agricultural Impacts on Biotic Integrity.** - There is a need to implement conservation practices that reduce agricultural runoff from surface and tile-drained waters and restore instream habitat in order to protect biotic integrity in freshwater watersheds. Forested riparian buffers and constructed wetlands may be the two most effective conservation measures to address this issue because they reduce multiple agricultural impacts by improving instream habitat quality, reducing nutrient loadings, ameliorating altered hydrology, and creating new wetland habitat for aquatic and migratory species.

**Metrics for measuring success of agricultural conservation practices**

*Recommendations.* - Multiple metrics have been used in bioassessment research; however, some indices appear to be more consistent than others in terms of their sensitivity to agricultural impacts and thus may be most useful in assessing the effects of conservation interventions in aquatic ecosystems (Table 1). Recommendations are based on published studies that tested the sensitivity of multiple metrics to agricultural impacts and those that used large-scale survey data to develop trends and conclusions. If practical, it would be best to include multiple indices, one each for macroinvertebrates, fishes, and habitat quality.

*Invertebrates.* - Among the studies that were reviewed for this summary, the Hilsenhoff Biotic Index (HBI) was the most consistent metric discriminating among the gradient of conditions associated with agricultural impacts on biotic integrity of aquatic macroinvertebrates. HBI is an abundance-weighted index that is computed based on the tolerance value of each macroinvertebrate taxon (Hilsenhoff 1987, 1988). The index values range from 0 to 10, with
higher values indicating more degraded water quality. In general, HBI values in the studies that were reviewed increased with increasing nutrient concentrations (Stone et al. 2005, Wang et al. 2007), decreased with increasing forested riparian buffer or area (Stone et al. 2005, Heatherly et al. 2007) and decreased with increasing habitat quality (Stone et al. 2005). One drawback to the wide application of this index, however, is that in most studies the HBI has been modified to better represent taxonomic compositions for individual states. Two different versions of HBI were used in two separate Illinois studies: a version of HBI calibrated for Illinois taxa called the macroinvertebrate index (MBI, Fitzpatrick et al. 2004) was used by Heatherly et al. (2007) for a statewide survey, and a version of HBI calibrated for Nebraska (NDEQ 1991) was used by Stone et al. (2005) for a watershed study conducted in southern Illinois. A general version of the HBI (Hilsenhoff 1987) or even a family level version (FBI, Hilsenhoff 1988) might be applicable for the broad-scale CEAP modeling. The HBI is commonly used to assess organic pollution in stream surveys and thus, data may be more readily available for modeling purposes. It should be noted that the purpose of the family level index (FBI) is to provide a rapid, but less critical, evaluation of streams and should not be used as a substitute for the HBI when detailed taxonomic information is available. A second macroinvertebrate metric that was consistent across those studies that reported taxonomic composition was the increased relative abundances of pollution tolerant taxa (i.e., % oligochaetes and non-insects) with increasing nutrients.

Several macroinvertebrate indices commonly used to evaluate biotic integrity appear to be too variable among studies to be useful in comparing environmental conditions among midwestern agricultural watersheds. The EPT metric did not accurately represent negative effects of increased nutrient loadings in agricultural watersheds in a southern Illinois because EPT taxa in these highly degraded systems were generally rare and the mayfly taxa that were present were pollution tolerant genera (Stone et al. 2005). Although it appears that in some cases agricultural watersheds in the Midwest may already be past the threshold where changes in EPT would be evident, other researchers have found the EPT index to be an accurate predictor of degradative impacts on biotic integrity in agricultural streams in Nebraska (While et al. 2000) and wadeable streams in Ohio (Miltner and Rankin 1998). Standard functional structure metrics (e.g., functional feeding groups) were also reported to be inappropriate for assessing agriculturally impacted streams (While et al. 2000, Stone et al. 2005), possibly related to the high degree of historical impacts and landuse change already enacted in these systems. Although
mussels are clearly negatively affected by agricultural impacts (e.g., Poole and Downing 2004), they are not likely to be useful for assessing short term changes in agricultural watersheds due to a long-term extinction debt in which local extinction of organisms with low dispersal potential continues to occur long after stream habitat and quality changes have been made (Tilman et al. 1994, Hanski and Ovaskainen 2002).

Fish.- Studies reviewed for this summary that assessed fish communities used some version of the Index of Biotic Integrity (IBI) developed by Karr (1981). The original IBI was developed for use in small warmwater streams in Illinois and Indiana and has been used by many researchers to assess warmwater streams throughout the central United States. As the IBI became more widely used, different versions were developed that modified those metrics that were insensitive to environmental degradation in a particular geographic region or stream type. Miltner and Rankin (1998) used a version of the original IBI that was modified for Ohio (Ohio EPA 1987, Yoder and Rankin 1995); whereas, Wang et al. (2007) used an IBI score computed using both coldwater (Lyons et al. 1996) and warmwater (Lyons 1992) versions, and used the higher of the two scores to assess fish assemblages in Wisconsin. Both studies found that the IBI was a sensitive metric to evaluate agricultural impacts on fish assemblages. Miltner and Rankin (1998) reported that IBI scores were negatively correlated with increasing concentrations of nitrogen and phosphorus in smaller streams (headwaters, wadeable) and provided some evidence for a direct link between total phosphorus and IBI scores. In Wisconsin streams, ecoregions with lower amounts of agriculture had higher fish IBI scores and fish IBI scores showed decreasing trends as nitrogen and phosphorus concentrations increased (Wang et al. 2007). A second fish metric that was consistent across these fish assessment studies was the decreased relative abundances or species numbers of sensitive fish taxa with increasing nutrients. If the SWAT model developed for CEAP is limited to the midwestern U.S., it may be that a more general version of IBI could be used each for warmwater and coldwater stream fish assemblages. Currently, almost all state Environmental Protection Agencies use some form of IBI (www.epa.gov), so the data needed to calculate IBI scores for many areas are likely available.

Habitat.- Most of the studies that were reviewed for this summary reported increased HBI and decreased IBI scores with increasing habitat degradation, as measured by either the QHEI or EPA Rapid Bioassessment. Although not a metric of biotic integrity, habitat assessment would provide important information on the environmental conditions influencing
the biotic metrics. Both Rapid Bioassessment and QHEI indices appear to be very good at assessing habitat quality in agricultural watersheds; however, for the purposes of the SWAT modeling for CEAP it would be recommended to use EPA’s Rapid Bioassessment for the reason that it is more commonly used in habitat surveys than the QHEI.

Summary of recommendations for CEAP modeling efforts

1. If practical, it would be best to include at least one index each for macroinvertebrates, fish, and habitat

2. Recommended indices:
   - **Fish**: IBI (versions that have been adjusted by state would be ideal, but if this is too impractical for large scale modeling it may be possible to just use two versions of IBI: one developed for warmwater systems, and one developed for coldwater systems)
   - **Macroinvertebrates**: HBI (general index or one calibrated by state); also an index based on the % oligochaete and non-insects might be useful
   - **Habitat**: EPA’s Rapid Bioassessment, QHEI: Both indices appear to be very good at assessing habitat quality in agricultural watersheds; however, for the purposes of the SWAT modeling for CEAP it would be recommended to use EPA’s Rapid Bioassessment for the reason that it is more commonly used in habitat surveys than the QHEI
References


Table 1. Summary of literature reviewed for biotic index use in SWAT modeling for CEAP.

<table>
<thead>
<tr>
<th>Organism</th>
<th>Scale (W, S)</th>
<th>Region</th>
<th>Landuse (Ag-T, Ag-NT)</th>
<th>Sampling</th>
<th>Results</th>
</tr>
</thead>
<tbody>
<tr>
<td>Macroinvertebrates</td>
<td>(17)</td>
<td>S W</td>
<td>S. IL</td>
<td>Ag-NT</td>
<td>1. Intensive (3 streams)</td>
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<td></td>
<td></td>
<td></td>
<td></td>
<td>(15 sites)</td>
<td>2. Rapid bioassessment</td>
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<tr>
<td></td>
<td></td>
<td></td>
<td></td>
<td>(15 watershed sites)</td>
<td>% noninsect taxa (esp. oligochaete) &amp; HBI sig. correlated with physical habitat score, % riparian forest, ORP - EPT, taxa richness, functional groups not reliable</td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td>Rangeland &amp; cropland</td>
<td>Ap, Jn, Au, Oc (1998)</td>
<td>% dominance &amp; EPT performed best (rather than HBI, CBI); functional groups not reliable</td>
</tr>
<tr>
<td></td>
<td>(4)</td>
<td>St</td>
<td>IL</td>
<td>Urban &amp; Ag</td>
<td>53 sites</td>
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<tr>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td>MBI (a version of HBI) values for Ag streams decreased as stream type went from forested to agricultural to urban</td>
</tr>
<tr>
<td></td>
<td>(18)</td>
<td>St</td>
<td>WI</td>
<td>Urban Forested &amp; Ag</td>
<td>240 wadeable streams 1st-4th order</td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td></td>
<td>WQ monthly My-Sp</td>
<td>EPT, HBI, mean tolerance values strongly correlated with nutrients</td>
</tr>
<tr>
<td></td>
<td>(11)</td>
<td>St</td>
<td>OH</td>
<td></td>
<td>523 sites across Ohio</td>
</tr>
<tr>
<td>Fish</td>
<td>(18)</td>
<td>St</td>
<td>WI</td>
<td>Urban Forested &amp; Ag</td>
<td>240 wadeable streams 1st-4th order</td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td></td>
<td>WQ monthly My-Sp</td>
<td>IBI; % carnivorous, intolerant, omnivorous; salmonid abundance correlated most with nutrient measures</td>
</tr>
<tr>
<td></td>
<td>(11)</td>
<td>St</td>
<td>OH</td>
<td></td>
<td>842 sites across Ohio</td>
</tr>
<tr>
<td>Mussels</td>
<td>(13)</td>
<td>St</td>
<td>IA</td>
<td>Ag</td>
<td>118 sites</td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td>Species richness declined w/ loss of wooded riparian habitat</td>
</tr>
</tbody>
</table>

S= Stream, St= statewide; W = watershed; Ag-T = Agricultural, tiled; Ag-NT = Agricultural, not tiled
Table 2. Citations for state modifications to the fish index of biotic integrity (IBI) and to Hilsenhoff’s biotic index (HBI) for macroinvertebrates.

Citations for HBI modifications by state:
2. Ohio (ICI): DeShon 1995
3. Illinois (HBI): used Nebraska DEQ (1991) tolerance values - because they were derived from similar, primarily agricultural, streams that are subjected to a similar array of disturbances and have similar taxonomic composition to Illinois.

Citations for IBI modifications by state:
2. Ohio: Ohio EPA 1987; Yoder and Rankin 1995
3. Wisconsin coldwater version (Lyons et al. 1996)
4. Wisconsin warmwater version (Lyons 1992)
5. Also see: www.epa.gov/bioindicators/html/ibi-metrics.html

Citations for habitat assessments:
1. QHEI
   a. Rankin 1995
   b. PDF version at: www.epa.state.oh.us/dsw/bioassess/BioCriteriaProtAqLife.html
2. Habitat Rapid Bioassessment: Barbour et al. 1999

Citations for Macroinvertebrate Rapid Bioassessments:
1. Barbour et al. 1999
Annotated literature summary of primary review papers

1. Stone et al. 2005 (Southern Illinois -Midwest, Non-tiled)

   **Site**: low gradient, substrate 14-55% silt, 26-51% sand, 17-58% gravel; dominated by runs and pools; watersheds 86-100% row crop

   **Objectives**
   (a) Characterize aquatic macroinvertebrate communities in streams
   (b) Identify riparian and instream factors influencing macroinvertebrate communities (i.e., biotic integrity).

   **Methods**
   (1) Sampled 15 headwater sites once; all had 15-m buffer zones comprised of exotic cool-season grasses and a variety of forbs and shrubs.
   (2) Used Rapid Bioassessment for physical habitat scoring (June) and macroinvertebrates (May) at 15 sites
   (3) Sampled 3 headwater sites intensively; characterized them as having riparian forest cover of low (6%), medium (22%), and high (31%).
   (4) At 3 sites, collected macroinvertebrates monthly
   (5) At 3 sites, collected BOM every 3 months m(CPOM, FPOM, VFPOM)
   (6) At 3 sites, temperature data collected every 2 hours at intensively monitored sites
   (7) Monthly nutrient analyses for all 15 sites during baseflow conditions (nitrate, ammonium, ORP)

   **Metrics**: Diversity (H'), % dominant taxon, richness, HBI, functional feeding groups, EPT Stream infrastructure: % forested riparian zone, substrate heterogeneity, BOM, bioassessment for physical habitat scoring (Barbour et al. 1999; range 0-100), nitrate N, ammonium N, ORP

   **Macroinvertebrates**: Intensive (monthly for 1 y at 3 streams); Rapid bioassessment on 15 sites for inverts and habitat (USEPA multi-habitat procedure for low gradient streams, Barbour et al. 1999)

   **Results**
   **Low riparian forest cover**
   1. Dominated by silt (significantly higher than medium or high forest cover)
   2. Significantly higher CPOM (also more corn and grass CPOM)
   3. Significantly higher macroinvertebrate densities (approximately 80% oligochaetes)
   4. Significantly higher densities and biomass of oligochaetes, bivalves (*Sphaerium*), ostracods
   5. Significantly higher densities and biomass of noninsects
   6. Significantly lower dipterans than medium stream, but not high stream
   7. Significantly higher % dominant taxon (87%) than in med (71%) and high (65%)
   8. Significantly lower diversity (0.4) than med or high (both 0.5)

   **Medium and high forest cover**
   1. Sand and gravel more dominant
2. Significantly higher insects (mostly dipterans, Chironomidae and Ceratopogonidae) in medium than low, but no significant differences between high and low
3. Significantly higher dipterans in medium than low, but no significant differences between high and low

General trends (not significant)
1. Higher densities and biomass of mayflies (Caenidae, Baetidae) and odonates (Libellula, Enallagma, Ischnura) in low cover stream
2. Higher densities and biomass of beetles (Hydrophilidae, Dytiscidae) in high cover stream
3. Higher densities and biomass of gastropods (Physella) in low stream
4. Trichoptera, Hemiptera, and Megaloptera very low at all sites
5. Collector gatherers dominated all sites (oligochaetes, chironomids)
6. Shredders absent from all sites
7. Filterers (mayflies) and scrapers higher at low cover site
8. Assemblages in all streams were heavily dominated by pollution-tolerant taxa (tubificid worms, fingernail clams, and pulmonate snails). Sphaerium are often found in degraded systems; Caenis, Baetis, and Callibaetis mayflies are relatively tolerant commonly found in degraded streams.

Indices
1. At the 3 intensive sites, only % dominant taxon and diversity showed any significant differences (% taxon higher at low cover site, diversity low at low cover site)
2. EPT tended to be higher at the low cover site (Caenidae, Baetidae)
3. At the 15 rapid bioassessment scale, only HBI showed significant correlation with stream integrity.
   a. HBI (range: 2.5-4.9; average = 3.5±0.2) significantly decreased with increased % forest buffer (range: 1-38%) and increased habitat score (range: 30-90)
   b. HBI significantly increased with increased ORP (range: 0.4-3.3 mg/L)
   c. The best relationship was Modified HBI scores and Physical habitat score ($r^2 = 0.72$, $P = 0.0001$)

Results summary
(1) Comparison of low, medium, and high riparian forest (3 streams)
   (a) Significantly higher silt, VFPOM, and CPOM in low-forested riparian cover stream than medium and high cover streams
   (b) Significantly greater abundances and biomass in low forested buffer stream, related to increased Oligochaetes, Sphaerium bivalves, and ostracods.
   (c) Increased mayflies at low forested buffer stream (Caenidae, Baetidae)
   (d) Low cover site had significantly higher greater % dominant taxon values than medium and high sites, and significantly lower Shannon diversity.
   (e) EPT was low and variable at all sites; (HBI range = 3.6-3.9)
   (f) % insect contribution to density increased with % riparian forest cover

(2) Comparison of rapid bioassessment sites (15 streams)
   (a) EPT richness low (range = 0 to 2); too was low and variable for meaningful statistics
(b) % dominant taxon and taxa richness showed no relationship with riparian vegetation, water chemistry, or physical habitat scores
(c) Modified HBI (range = 2.5-4.9; average = 3.5±0.2); significantly related to ORP (r²=0.63, P = 0.0004), % riparian forest cover (r²=0.61, P = 0.0006), and instream physical habitat scores (r²=0.72, P < 0.0001).
(d) Multiple regression analysis showed that instream physical habitat was the most important variable related to HBI.

Conclusions
(1) HBI was a good predictor of agricultural impacts related to phosphorus (ORP, range ~0.3-3.5 mg/L), instream habitat, and % riparian forest; HBI was the most useful metric for discriminating among gradient conditions.
(2) Taxa richness and EPT not a good predictors
(3) Although stream communities in all sites were dominated by pollution tolerant taxa (worms, snails, fingernail clams), there were differences in communities across a gradient of conditions.
(4) Of all the many human impacts of these streams, sedimentation was the most obvious and likely the most important.
(5) There is potential for bioassessment in highly degraded agricultural streams, and the USEPA’s Rapid Bioassessment Protocols are useful.

2. Whiles et al. 2000 (NE Nebraska – Northern Plains)
   Site: 6 study reaches in the Willow Creek watershed; sites were low gradient, substrate 65-85% sand; dominated by runs and pools; minimal canopy cover

Objectives
(a) Use standard invertebrate rapid bioassessment methods based on those outlined in US EPA in an attempt to identify stream reaches contributing to water quality degradation in NE Nebraska reservoir

Questions
(a) Are standard invertebrate bioassessment methods effective for discriminating among differentially impacted, low-gradient streams draining an intensely agricultural region of the Northern Plains?
(b) Which individual metrics will perform best in this region?
(c) Will seasonal patterns of bioassessment scores differ from those of other regions?
(d) What variables will most influence bioassessment results, and thus aquatic ecosystem health?

Methods
1. Measured: nutrients (TN, NO₃-N, NH₃-N, TP, ORP, Cl⁻) and TSS during non-freezing months (May-Sept 1996; April-Oct 1997); also collected DO, Cond, pH, Temperature, Q
2. Metrics: Modified HBI, CBI (developed by NDEQ)
3. Stream infrastructure: Habitat quality (protocols for low gradient streams, Barbour et al. 1997) – performed once each year in August
5. Designated one site as a reference site

Notes: (1) These sites were dominated by sandy substrates 65-85% of substrate composition, as opposed to silt and gravel like the Illinois sites.

Results
1. Site with high nutrients and low habitat had low CBI scores (site 1)
2. Site with low nutrients and high habitat had low CBI scores (site 5)
3. % dominance showed the greatest discrimination between ref site and sites 1 and 5, followed by EPT
4. HBI and taxa richness metrics not as discriminatory but highest precision

Conclusions
(1) US EPA Rapid Bioassessment are suitable for preliminary identification of relative impacts on stream reaches in NE Nebraska region
(2) Low biotic integrity was associated with (a) high nutrients even with high habitat quality and (b) low nutrients and degraded (channelized) habitat
(3) Functional group metrics were unreliable
(4) % dominance and EPT metrics performed best for identification of impacted stream reaches
(5) HBI worked, just not as effectively as % dominance or EPT
(6) Stresses importance of riparian zones and that similar importance should be given to prairie streams as forested streams
(7) Landscape-level variables were the most accurate predictors of stream biotic integrity

3. Heatherly et al. 2007 (IL, statewide survey: urban & agricultural; tiled & not tiled)
   Sites: 53 wadeable streams across Illinois

Objectives
(a) Identify stressors that most strongly influence the biotic integrity of stream macroinvertebrate communities
(b) Determine what stressors most influence the macroinvertebrate communities of specific stream types

Hypotheses
(a) The degree of nutrient enrichment and the amounts and types of habitat degradation would vary across Illinois in relation to land use, and would be reflected in the macroinvertebrate community
(b) Physical habitat quality would be the most important factor controlling biotic integrity among streams, across all sites

Methods
(1) Chemical data: NO$_3$-N, NH$_3$-N, NH$_4$-N, DRP, TN, TP, DOC, Chl $a$, DO, Temp, pH, Specific conductivity
(2) Habitat: (a) Substrate composition (b) Rapid Bioassessment (Barbour et al. 1999)
(3) Macroinvertebrates: USEPA multihabitat Rapid Bioassessment for wadeable streams (Barbour et al. 1999)
(4) Used Macroinvertebrate Biotic Index (MBI), which is a version of the HBI that has been calibrated for Illinois taxa (Fitzpatrick et al. 2004).

**Results:** Highest biotic integrity in forested streams, lowest in urban streams
(1) Samples from 2 highly impacted streams in industrial areas only had 42 and 53 macroinvertebrates in samples collected
(2) Richness ranged from 7 to 29 across all study streams
(3) Identified 4 groups of streams (used cluster analysis, PC-ORD):
   - **Group 1:** mostly agricultural
     (a) Relatively higher taxa richness and diversity (than Grp 3 & 4; than Grp 4)
     (b) Lower MBI (than Grp 4)
     (c) % Oligochaeta higher (than Grp 2)
     (d) Higher EPT (than Grp 3 & 4)
   - **Group 2:** mostly forested (primarily southern IL sites)
     (a) Lowest nutrients and dissolved ions
     (b) Relatively higher taxa richness and diversity (than Grp 3 & 4; than Grp 4)
     (c) Lower MBI (than Grp 3 & 4)
     (d) Lowest % Oligochaeta
     (e) Higher EPT (than Grp 3 & 4)
   - **Group 3:** mostly urban
     (a) Highest nutrients and dissolved ions
     (b) Much higher DRP than other groups
     (c) Relatively lower taxa richness (than Grp 1 & 2)
     (d) % Oligochaeta higher (than Grp 2)
     (e) Lower EPT (than Grp 1 & 2)
   - **Group 4:** 2 highly degraded urban streams
     (a) Relatively lower taxa richness and diversity (than Grp 1 & 2)
     (b) % Oligochaeta higher (than Grp 1 & 3)
     (c) Lower EPT (than Grp 1 & 2)

(4) Forested and agricultural streams aligned best with habitat quality and stable substrata
(5) Urban streams aligned best with higher nutrient and dissolved ion concentrations

**Conclusions**
(1) Unable to support hypothesis that habitat quality would be the primary factor governing biotic integrity because habitat degradation was generally evident in streams with elevated nutrient concentrations.
(2) Analyses indicate that habitat quality and nutrient concentrations were of equal importance in structuring macroinvertebrate communities across Illinois streams.
(3) Inverse relationship between habitat quality and nutrient concentrations
(4) Both N and P were identified as predictors of macroinvertebrate assemblages, but in many Illinois streams, N concentrations are sufficiently high to preclude N limitation of algae – Heatherly et al. suggest that P may be the critical nutrient affect biotic integrity among the sites, particularly in agricultural regions of the state.

(5) Stream macroinvertebrate richness and diversity was lowest in streams draining urban areas (runoff from impervious areas, channelization) and highest in forested areas of southern IL.

(6) Agricultural areas typically were dominated by chironomids and oligochaetes

4. Wang et al. 2007 (WI, range of anthropogenic influence)  
   Sites: 240 wadeable streams across Wisconsin (1rst to 4rth order)

Objectives
   (a) Determine how macroinvertebrate and fish assemblage measures correlated with different forms of nutrients (N, P)  
   (b) Examine the relationships between key biological measures and nutrient forms to quantify potential threshold concentrations for nutrients beyond which macroinvertebrate and fish assemblages are likely to be substantially degraded  
   (c) Evaluate the importance of nutrient concentrations in influencing macroinvertebrate and fish assemblages relative to other physicochemical variables at different spatial scales

Methods
   (a) Four ecoregions: Northern Lakes and Forest (NLF), North Central Hardwood Forest (NCHF), Driftless Area (DFA), Southeast Wisconsin Till Plains (SWTP).  
   (b) Digitized land use and land cover  
   (c) Water quality and discharge measurements collected May - September  
   (d) TP, DP, TKN, NO3-N, NH4-N (TN=TKN+NO3-N)  
   (e) Fish (35 assembly measures, IBIs for WI (a) coldwater and (b) warmwater streams), Habitat (WI Forest Service Methods)  
   (f) Macroinvertebrate (HBI, MIBI, 24 summary measures) – MIBI is a macroinvertebrate index developed for Wisconsin

Results
   (a) NLF (highest forest cover) had lowest and SWTP (highest urban and agricultural uses; lowest forest cover) had highest median concentrations of TP and TN  
   (b) HBI ranged from 1 to 9 (mean = 5)  
   (c) IBI ranged from 0 to 100 (mean = 40)  
   (d) Medians of TP, TKN, TN, NH4-N explained most of the variances in fish variables and correlated with the most macroinvertebrate measures

Conclusions
   (a) Nutrients have direct or indirect links to biological assemblages  
   (b) Wedge-shaped relationship between nutrients and biota indicate that at low nutrient concentrations, factors other than nutrients are predominant factors limiting health of
biological assemblages, whereas at high nutrient concentrations, nutrients may be the predominant factors affecting biological assemblages.
(c) Nutrients by themselves had relatively small direct influence on biological assemblages in wadeable streams; however, nutrient concentrations and the interactions between nutrients and other environmental variables explained more that half of the variation in fish and macroinvertebrate assemblages. Results imply that nutrients are among the key factors in determining the biological health in the studied streams.

(d) 5. Miltner and Rankin 1998 (Ohio –Midwest, unknown % agriculture versus urban; unknown tiled versus untiled – but this study included many representative sites across the state)

Sites: streams and rivers throughout Ohio (842 sites for fish analyses, 523 sites for macroinvertebrate)

Objectives
(a) Determine relationship between primary nutrients and fish or macroinvertebrates
(b) If relationship exists, estimate variation explained by nutrients (N, P) versus other variables, especially habitat
(c) Quantify potential threshold levels of nutrients in streams beyond which fish and macroinvertebrate community structure is likely to significantly degraded
(d) Investigate relationship between macroinvertebrate and fish community structure and increasing nutrient concentrations

Methods
(1) Used Ohio EPA data (biological, physical, chemical)
(2) Multiple regression model to determine relationship of IBI and ICI with: TIN, TP, QHEI, and TIN*TP for 4 stream size classes (headwaters, wadeable, small rivers, larger rivers)
(3) ANCOVA, MANCOVA
(4) Headwaters (<51.8 km2); Wadeable streams (51.8<518.1 km2); small rivers (518.1<2590.7 km2); larger rivers (≥2590.7 km2)

Metrics: QHEI (Habitat); IBI (modified for Ohio); ICI (Macroinvertebrate Community Index, DeShon 1995);

Results
Habitat
(1) Generally explained 20-30% of the variation in IBI or ICI scores across stream size and models, demonstrating the importance of habitat

Nutrients
(1) TP explained 2-16%, depending on stream size and model.
(2) IBI and ICI scores negatively correlated with increasing nutrient concentrations (TP, TIN) for small streams (headwaters, wadeable)
(3) IBI scores negatively correlated with increasing TIN concentrations in small rivers
(4) No correlation between IBI and nutrients in larger rivers
Fish
(1) Significantly higher IBI scores in headwater streams with TIN < 1.37 mg L$^{-1}$ and P < 0.17 mg L$^{-1}$.
(2) Significantly higher IBI scores in wadeable streams with decreasing TIN and P
(3) IBI scores were ill-defined or not related to nutrients in small and large rivers

Invertebrates
(1) ICI scores were less conclusive than IBI, except for wadeable streams, which showed decreased ICI with increased nutrients

Conclusions
(1) Biotic integrity for fish and invertebrates in rivers and streams is negatively correlated with increasing nutrient concentration, especially phosphorus (range 0.06 (25$^{th}$ percentile) to 1.70 (90$^{th}$ percentile))

(2) Effects of nutrients on fish and invertebrate biotic integrity are most evident in low- to mid-order streams (headwater, wadeable, and some small river)
- Pollution sensitive fish (mostly specialized insectivores), insectivores, and top carnivores showed a negative relationship to increasing nutrient concentration in headwaters, wadeable streams and small rivers
- Decline in the number of sensitive fish species showed strongest correlation to increased nutrient concentrations
- Relative abundance of top carnivores decreased with increasing nutrient concentrations in wadeable streams and small rivers; insectivores decreased with increasing nutrient concentrations in headwaters and wadeable streams
- Relative abundance of tolerant and omnivorous fishes increased significantly in relation to nutrient enrichment in headwaters, wadeable streams, and small rivers; highest relative abundances evident at intermediate levels of enrichment, especially in wadeable streams
- IBI and ICI scores negatively correlated with increasing nutrient concentrations (TP, TIN) for smaller streams (headwaters, wadeable)
- IBI scores negatively correlated with increasing TIN concentrations in small rivers

(3) Effects of nutrients were more obvious on fish than invertebrates
- Number of EPT taxa and relative abundances of Tanytarsini midges decreased with increasing nutrients in wadeable streams
- Other dipterans and non-insects were positively associated with increasing nutrient concentration
- Increased mayflies at intermediate nutrient levels and scrapers at higher nutrient levels may have offset ICI metric scoring

(4) Habitat quality, as measured by QHEI, is an important factor related to biotic integrity
- Habitat generally explained 20-30% of the variation in IBI or ICI scores across stream size and models
6. Poole, E. K., and J. A. Downing (2004) (IA, tiled or not tiled, % agricultural)
   Sites: 118 sites

**Objectives**
(a) Quantify change in species richness between 1984-85 and 1998
(b) Determine relationships between site-specific, stream-reach characteristics, and changes in
   species richness
(c) Analyze relationships between current watershed characteristics and rate of change in
   species richness at the watershed scale

**Methods**
(a) Stream surveys for mussels
(b) Water quality (TN, TP, TSS, alkalinity) and habitat assessments at 5 transects/sampling
   site (stream shading, % substrate type, stream depth and width, riparian composition)
(c) Classified % landuse and geology (GIS)

**Results**
(a) Average mussel species richness declined by >50% between 1984-85 and 1998
(b) Increase from 6% of sites with no living mussels (1984-85) to 47% of sites with no living
   mussels (1998) – Note that these sites were chosen to represent the region’s least degraded
   mussel habitat
(c) 58% of the sites lost >75% of the species richness in 10 years

**Conclusions**
(a) Physical habitat degradation and nutrient pollution are important and confounded
determinants of biotic integrity in Illinois streams