Technical Support Document for Nutrient Water Quality Standards for Ohio Rivers and Streams

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1. Introduction

The stated objective the Federal Water Pollution Control Act, commonly known as the Clean Water Act (CWA), is to restore and maintain the chemical, physical, and biological integrity of the Nation's waters. Progress toward that objective has been remarkable in light of the existing conditions at the time of the Act's initial passage in 1972. Prior to 1985, most of Ohio's surface waters failed the basic goal of fishable and swimmable, with 20% of the waters showing evidence of either acute or chronic toxicity due to egregious levels of pollution from industrial sources and under-treated municipal sewage. Presently, the majority of Ohio's surface waters meet the basic CWA goal, and toxicity is rarely observed. Furthermore, many rare and pollution-sensitive fish species that were restricted to a handful of refugia prior to 1985 have recovered much of their historic range in the state. This dramatic recovery was the direct result of an investment in wastewater infrastructure combined with statutory authority to set pollution limits. Where impairment persists, most can be attributed to poor habitat quality, followed by excessive sediment, organic enrichment, and nutrient enrichment. Frequently, these causes are intermingled.

For example, organic enrichment and nutrient enrichment are often two sides of the same coin. Organic matter from under- or untreated sewage is rich in phosphorus and nitrogen, and those nutrients can be liberated during decomposition. Conversely, excessive algal growth fueled by sources of inorganic (e.g., agricultural fertilizers) or mineralized (e.g., treated municipal effluent) nutrients can cause organic enrichment if the algae senesce and decay. Sediment loads from overland and stream bank erosion carry phosphorus, as phosphorus adsorbs to clay particles, and that phosphorus can be liberated to the water column if the sediments become anoxic (Surridge et al. 2007, Nguyen and Sukias 2002), over-taxed (Koopmans et al. 2004, Nguyen and Sukias 2002), or by enzymatic activity of the microbial community (Marxsen and Schmidt 1993). Finally, habitat quality of the stream channel and riparian zone influences both nutrient and sediment retention and assimilation, adding complexity to the outcomes of nutrient enrichment (Munn and Meyer 1990, Naiman et al. 1988, Malanson 1993; and see Ohio EPA 1999 for a literature review on the subject).

A common thread running through the principal remaining causes of impairment is that their sources are largely diffuse and primarily associated with agriculture. The diffuse nature of the sources is one of the reasons[1] that the associated impairments have not been remediated. Quite simply, most of the regulatory infrastructure is designed to handle pollutants emanating from point sources that can be either modeled mathematically, or have reasonably well-defined dose-response curves. That said, a considerable intellectual infrastructure has been cultivated to assess, quantify, and model pollutants and pollution loads associated with diffuse sources via the impetus to complete Total Maximum Daily Load (TMDL) studies. Were the regulatory and statutory infrastructure to follow suit, implementation of water quality standards (WQS) to address these causes would be relatively straightforward—at least within the confines of the experience accrued in developing TMDLs to address diffuse sources, but not necessarily in terms of the historic regulatory structure. Defining water quality criteria to support a standard for nutrients, however, has proven to be anything but straightforward.

Nutrients, by definition, are a necessary component of living things and ecosystems, such that the first practical limitation to defining a criterion, is determining how much of a natural and necessary component is too much. One might be tempted to conclude that any amount greater than that

[1] Another reason for lingering impairment is that local drainage laws supersede the CWA, and the drainage practices so sanctioned are completely at odds with the objective of the CWA.

necessary to sustain the system is too much. This line of reasoning girds the reference approach, wherein statistical distributions of nutrient concentrations measured (or modeled, e.g., Soranno et al. 2008) from a collection of least-disturbed to near pristine sites defines the upper limit of acceptable concentrations. Of course, ambient nutrient concentrations measured from working landscapes are higher than the upper bounds of reference distributions, and little or no association with biological condition, as measured by fish and macroinvertebrate communities, is observed until ambient concentrations exceed reference concentrations by two to six times (Weigel and Robertson 2007), to an order of magnitude (Miltner and Rankin 1998). Alternately, one might reasonably conclude that the point where demonstrable harm is caused by an excess of nutrients is an obvious start. Most WQS are predicated on this line of reasoning, and they are usually supported by laboratory toxicity tests. Nutrients, however, are generally not toxic at concentrations typically measured in the field, thus obviating the laboratory approach. The work-around, as alluded to, is to derive nutrient criteria based on field studies (e.g., Heiskary and Markus 2003, Camargo et al. 2005b, Skoulikidis et al. 2004, Donohuea et al. 2006, Ponader et al. 2007, Smith et al. 2007, Wang et al. 2007, Soranno et al. 2008, Miltner 2010).

Defining nutrient thresholds based on empirical field studies, however, runs up against practical limitations having to do with the dynamic nature of streams, not least of which is that the nutrient-eutrophication relationship is itself complex (Biggs 2000, Munn et al. 2010). Also, the impact of eutrophication on higher trophic levels is difficult to quantify because fish and macroinvertebrate communities are strongly influenced by physical habitat (Miltner and Rankin 1998, Wang et al. 2007; Munn et al. 2010), flow regime (Poff and Allen 1995), geomorphic condition (Walters et al. 2003, Mazeika et al. 2006) and landscape factors (Passy et al. 2004). Fortunately, the relationships between nutrients and stream eutrophication have been well documented (Dodds et al. 1997, Smith et al. 1999, Biggs 2000), and a sufficient number of field studies exist that trace links between nutrients and algae, macroinvertebrates, or fish, such that a reasonably complete picture emerges of how biological condition changes over a nutrient gradient. This understanding helps resolve the implied tension between the amount of excess defined by the reference approach, and that defined by overt biological impairment. To see how this is so, a brief thumbnail sketch of the emergent picture is in order.

Laboratory (Hill and Fanta 2008) and mesocosm (Bothwell 1989) studies have shown that algal growth rates can be saturated at low concentrations of nutrients (P ~ 0.002–0.02 mg/l), and field studies have corroborated those findings (Biggs 2000). In the environment, however, saturating concentrations are functionally higher due to physical factors such as shading (Larned and Santos 2000) and scouring (Biggs 2000), and biological factors including grazing (Bourassa and Cattaneo 1998) and competition (Scott et al. 2008). Operationally, this translates into algae in oligotrophic streams being relatively more susceptible to changes in nutrient concentrations compared to meso- or eutrophic streams (Bowman et al. 2007). See Table 1 for a summary of studies reporting nutrient concentrations saturating to algal growth. However, field studies that look directly at the relationship between nutrient concentrations and algal composition or abundance suggest a response over a 3-order magnitude range of nutrient concentrations (see Table 2 for a summary), and demonstrate that changes occur across the continuum of stream trophic status (Hillebrand 2002). Studies examining how macroinvertebrate or fish assemblages vary over gradients of nutrient enrichment (see Table 3) similarly demonstrate, either directly or inferentially, response across the spectrum, though the responses are not always direct.

Table 1. Nutrient concentrations saturating to algal growth reported in the literature.

Study	Nitrogen	Phosphorus	Comments
Biggs (2000)	DIN 0.02	SRP 0.002	Concentrations saturating to growth
Bothwell (1985)		SRP 0.004	Concentration saturating to growth
Hill and Fanta (2008)		SRP 0.022-0.082	Experimental, concentration range saturating algal growth
Hill et al. (2009)		SRP 0.025	Concentration saturating algal growth; laboratory mesocosm
Larned and Santos (2000)		DP 0.004-0.070	Experimental manipulation, P given at saturating concentrations above the listed background concentrations resulted in increased algal abundance when light was available, low order, tropical streams
Rier and Stevenson (2006)	DIN 0.086	SRP 0.016	Concentrations saturating algal growth, streamside mesocosms (KY)
Rosemond et al. (2002)		SRP 0.025 - 0.05	Concentrations saturating heterotrophic pathway; field & experimental manipulation; headwater streams (CRC)

Table 2. Threshold nutrient concentrations or ranges where changes in algal biomass or assemblage structure occur as noted in the literature.

Study	Nitrogen	Phosphorus	Comments
Camargo et al. (2005b)	DIN 0.687 - 2.958	ortho-P 0.017 - 0.073	Empirical, increase in benthic chl-a in response to nutrient addition over range shown (Spain), large, oligotrophic rivers
Johnson and Hering (2009)		TP 0.05	Associations based on field observations, change in assemblage composition in response to TP; distinct threshold for algal assemblage. Northern EU, wadeable
Justus et al. (2010)	TN 0.4	TP 0.018	Association of biotic index for algae and nutrient concentrations; (AK), oligotrophic, wadeable streams
Smith and Tran (2010)	TN 0.7	TP 0.03	Protection of benthic algal communities in large (NY) rivers; CPA, Percentile, Cluster Analysis
Black et al. (2011)	TN 0.59 - 1.79	TP 0.03 - 0.28	Piecewise regression, algal metrics, small to large streams (USA)
Bowes et al. (2007)		SRP 0.04 - 0.09	Forced control of benthic algae by reducing concentrations to noted range, stream-side mesocosm study (GB)
Bowman et al. (2007)		TP 0.004 - 0.028	Empirical, increase in benthic chl-a in response to nutrient addition over range shown (Alberta), large, oligotrophic rivers
Carr et al. (2005)	TN 1.0	TP 0.03	CP in LOWESS, periphytic biomass (Ontario and Quebec), rivers and streams

Study	Nitrogen	Phosphorus	Comments
Cattaneo et al. (1997)	TN 0.330 - 0.507	TP 0.013 - 0.042	Empirical, increase in benthic chl-a in response to nutrient addition over range shown (Ontario & Quebec), wadeable, oligotrophic
Chambers et al. (2009)	TN 0.41 - 1.15	TP 0.012 - 0.101	Values listed are ranges of "Idea Performance Standards" to protect ecological condition in Canadian streams
Dodds et al. (1997)	TN 0.35	TP 0.03	Regression, levels chosen to prevent nuisance growths
Dodds et al. (2002, 2006)	TN 367-602	TP 0.027 - 0.062	CPA, field data (North American Rivers)
Kelly et al. (2008)	NO3 4.0	SRP 0.03	Empirical, diatom trophic index (UK), small streams
King (2009)	DIN 0.021 - 3.264 (0.634)	TP 0.018 - 0.555 (0.25)	CPA, algal metrics, experimental and observational studies (TX), wadeable
Lewis and McCutchan (2010)	DIN 0.30	TP 0.03	Empirical, no association between algal abundance and nutrients in oligotrophic streams, i.e., concentrations were less than those listed(CO), wadeable and small rivers
Lohman et al. (1992)	TN 0.244 - 1.782	TP 0.015 - 0.634	Maximum accrual of bethic algae higher at enriched sites (listed upper range value) compared to unenriched sites (lower listed value). Wadeable streams (MO).
Munn et al. (2010)		TP ~ 0.10	Linear response in benthic chl-a at TP concentrations less than 0.1 mg/l, flat response > 0.1 mg/l. (North American rivers)
Ponander et al. (2007)	TN 0.7, 1.5	TP 0.025, 0.075	Community shift in diatom composition (NJ), upland streams
Ponander et al. (2008)		TP 0.075	Community shift in diatom composition (NJ), coastal plain streams
Stevenson et al. (2008)		TP 0.01 - 0.02	CPA chl-a, algal metrics (Mid Atlantic)
Stevenson et al. (2006)	TN 1.0	TP 0.03	Association, limit cladophora cover (KY, MI), wadeable streams

Table 3. Threshold concentrations or ranges over which detectable changes occur in invertebrate and fish assemblages as reported in the literature.

Study	Invertebrates		Fish		Comments
	Nitrogen	Phosphorus	Nitrogen	Phosphorus	
Camargo et al. (2005a)	NO3 2.0				Toxicity to sensitive invertebrates
Camargo et al. (2005b)	DIN 0.687 - 2.958	ortho-P 0.017 - 0.073			Empirical, increase in invertebrates in response to nutrient addition over range shown (Spain), large, oligotrophic rivers
Evans-White et al. (2009)	TN 1.34	TP 0.06			CPA of field data (NE, MO, KS) small stream - large rivers
Heiskary et al. (2010)				TP 0.055, 0.10, 0.150	Protective levels based on empirical relationships with water quality parameters and biological metrics
Johnson and Hering (2009)		TP 0.1 ~ 1.0		TP 0.1 ~ 1.0	Associations based on field observations, change in assemblage composition in response to TP; changes detectable over range of concentration for fish and inverts. Northern EU, wadeable
Justus et al. (2010)	TN 2.0	TP 0.045	TN 0.5	TP 0.025	Values listed for inverts and fish implied from graphs and regression tree cuts from data in appendix (AK), oligotrophic, wadeable streams
King and Richardson (2003)		TP 0.017			CPA, macroinvertebrates (FL), Everglades
Meador and Carlisle (2007)			NOx 0.97-1.74	TP 0.09 - 0.21	Differences in mean concentrations for fish grouped as tolerant, moderate, intolerant
Newall and Tiller (2002)	TN 0.015 - 0.9	TP 0.02 - 0.04			Reference approach informed by biomonitoring, (AU - Victoria) small to large streams
Riva-Murray et al. (2002)	TN 0.05 - 2.24	ortho P 0.01 - 0.12			Macroinvertebrate assemblage structure differs over range of concentration, wadeable streams (NY)
Sheeder and Evans (2004)	DIN 2.01	TP 0.07	DIN 2.01	TP 0.07	Association, prevent biological impairment (PA) wadeable streams
Smith and Tran (2010)	TN 0.7	TP 0.03			Protection of invertebrate communities in large (NY) rivers; CPA, Percentile, Cluster Analysis
Smith et al. (2007)	NOx 0.98	TP 0.065			Respective community response thresholds, wadeable streams (NY)
Wagenhoff et al. (2011)	DIN 0.107 - 0.144				Initial decline in mayfly taxon, %EPT
Wang et al. (2007)	TN 0.85 - 1.68	TP 0.04 - 0.09	TN 0.54 - 1.83	TP 0.06 - 0.09	CPA of field data (WI) wadeable streams
Wiegle and Robertson (2007)	TN 0.634 - 1.925	TP 0.064 - 0.150	TN 0.634	TP 0.091 - 0.139	CPA of field data (WI) large rivers

For example, nutrient amendments to an arctic stream initially produced direct responses of increased production of algae, macroinvertebrate, and fish growth rates (Deegan and Peterson 1992, Peterson et al. 1993), followed by indirect responses in the macroinvertebrate community mediated by changes in composition of the primary producers. Another example of an indirect pathway was observed in a shaded first order stream in North Carolina where nutrient additions increased abundance and production of both macroinvertebrate primary and secondary consumers via a heterotrophic path (Cross et al. 2006). Lastly, King and Richardson (2007) working in the Everglades experimentally demonstrated that some macroinvertebrate taxa show a subsidy-stress response to nutrient enrichment, wherein abundance is initially stimulated at modest levels of enrichment, but lowered at high levels of enrichment due to competitive interactions between primary producers. Note the similarity to the artic stream example.

As fish are typically one to several trophic steps removed from primary producers, the effects of nutrient enrichment on fish are less direct, being mostly manifest through the influence of enrichment on dissolved oxygen (DO) concentrations, and especially on DO fluctuations. Sabater et al. (2000) observed a difference between daytime and nighttime DO concentrations of over 10 mg/l at an enriched site where benthic chlorophyll levels exceeded 500 mg/m^2, and reported that short episodes of hypoxia associated with wide DO swings were responsible for fish kills in the study area. In a study of large Minnesota rivers (i.e., >2600 km^2) the daily range of DO concentrations was correlated with total phosphorus and sestonic chlorophyll-*a* (Heiskary and Markus 2003), and, in turn, fish IBIs were poorer at sites with high maximum DO and wide DO swings, but showed no relationship with minimum DO. Miltner (2010) found that minimum daily DO concentrations were negatively associated with daily DO range, and that fish Index of Biotic Integrity (IBI) scores, after accounting for coldwater streams, were poorest at site with the lowest minimum DO concentrations.

Other studies have inferentially demonstrated negative effects of nutrient enrichment on macroinvertebrates or fish through direct gradient analysis (Carlisle et al. 2007, Smith et al. 2007, Meador and Carlisle 2007, Haase and Nolte 2008), associations with biotic indices (Miltner and Rankin 1998, Hering et al. 2006, Wang et al. 2007), or multivariate approaches including discriminant analysis (Norton et al. 2000) and canonical correspondence analysis (Riva-Murray et al. 2002). Although cause and effect was not directly demonstrated by these studies, eight of the studies partitioned the variance in biological response over several or more environmental gradients (i.e., land use, physical stream habitat quality, sediment, and water chemistry) in addition to a nutrient gradient, thereby building a circumstantial case for a causal link between nutrients and the biological response.

Taken collectively, the body of work outlined thus far demonstrates that biological communities in rivers and streams show response over a wide nutrient gradient. A summary plot of concentrations presented in Tables 1–3 further suggests that algal, macroinvertebrate, and fish assemblages all tend to show response over similar ranges of nutrient concentrations, especially with respect to nitrogen (Figure 1). With respect to phosphorus, fish show a response at higher concentrations compared to algae, and marginally higher compared to macroinvertebrates. The upshot of all this, is that there is a dose-response relationship of sorts, though that response cannot be interpreted in the traditional sense because of the indirect pathways over which it is expressed, and because of the confounding factors that tend to mute, obscure, or exacerbate the responses. The dose-response relationship, such as it is, can be exploited, however, because there is a reasonably predictable and consistent response between increasing nutrient concentrations and periphyton (reviewed by Hillebrand 2002), and between periphyton and DO concentrations (Morgan et al. 2006, Huggins and Anderson 2005, Heiskary et al. 2010, Miltner 2010). The Ohio EPA nutrient criteria study (Miltner 2010) was predicated on tracing the steps from nutrients to periphyton (as given by chlorophyll-*a*), from periphyton to DO, and from DO to macroinvertebrates and fish, with the goal of identifying benchmarks or thresholds at each step that would help define where a given water body is positioned along a continuum of enrichment.

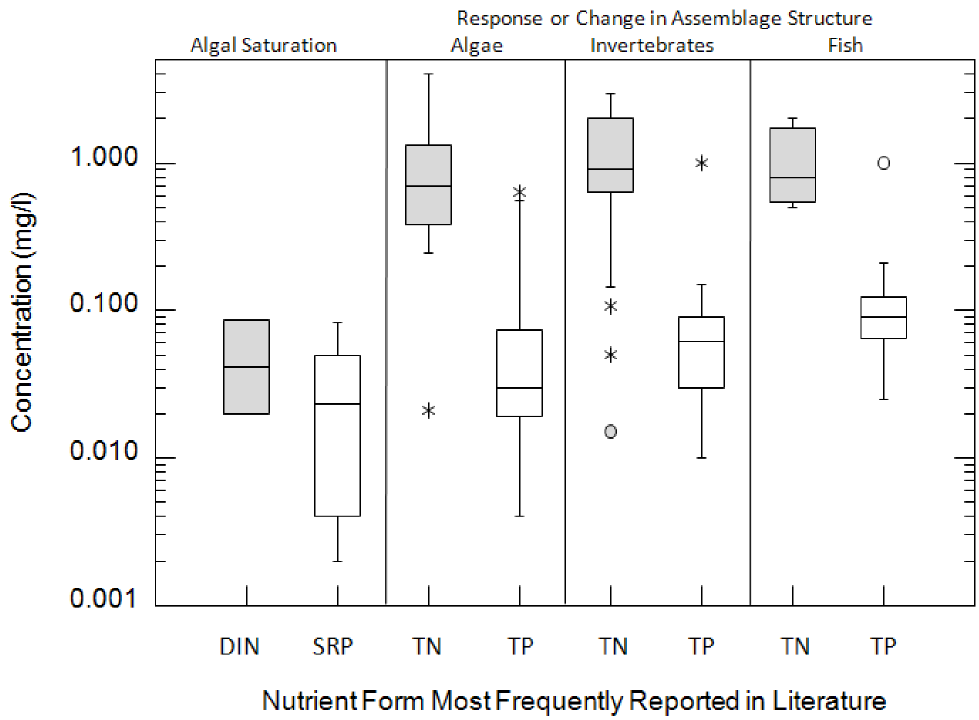

Figure 1. Distributions of data values presented in Tables 1, 2 and 3 plotted by taxonomic group. The distributions on the left end are for concentrations saturating to algal growth from Table 1. The distributions under the heading "Response or Change in Assemblage Structure" are from Table 2 for algae, and Table 3 for invertebrates and fish. All forms of a given nutrient were included the distributions, but the most frequently listed concentrations are noted on the x-axis.

2. Overview of the Ohio EPA Nutrient Study

2.1 Methods

The study area description and methods presented here are taken from (Miltner 2010) and from the Mutually Agreed Upon Nutrient Criteria Development Plan (http://water.epa.gov/scitech/swguidance/standards/criteria/nutrients/upload/Ohio-Outline-of-Methodology-to-Establish-Scientifically-Defensible-Nutrient-Water-Quality-Standards.pdf)

2.1.1 Study Area

One hundred and nine survey sites were selected to establish a gradient of anthropogenic enrichment and habitat quality based on a combination of historic water quality and stream habitat data, proximity to municipal wastewater plants, and land use from satellite imagery. Land use for each sampling location was derived from 30 meter resolution Landsat Thematic Mapper satellite imagery (September–October 1994) of land cover provided by the Ohio Department of Natural Resources. The percent of land area in the satellite data classed as urban or agricultural for the drainage upstream from each sampling point was used as an indicator of potential enrichment. For 19 sites that were situated on large rivers (i.e., an administrative demarcation for streams with drainage areas > 1300 km^2), the delineation of drainage land use upstream from a sampling point included all the area of principal tributaries up to a maximum area of 775 km^2 (i.e., one half the drainage area of the smallest large river site). Drainage area and local stream gradient were calculated for each site. Figure 2 shows the location of sites in Ohio, a frequency distribution of site drainage areas, and quantile plots of the percentage of urban and agricultural land in the upstream drainage for the sampling points.

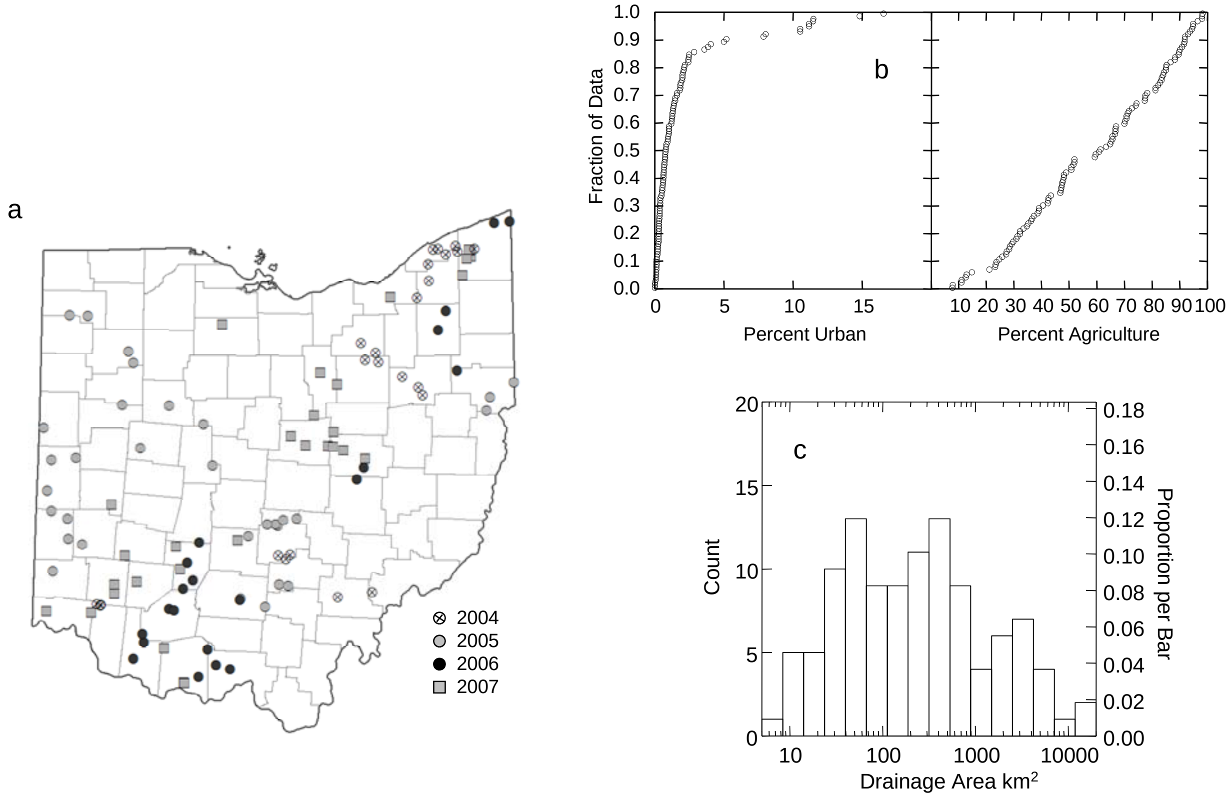

Figure 2. a) Sampling locations by year, b) cumulative distributions of percent urban and agricultural land uses upstream from sampling locations, and c) the frequency distribution of drainage areas at sampling locations.

2.1.2 Chemical, Biological, and Physical Sampling

Sites were sampled in batches between 2004 and 2007 such that roughly one quarter of the sites were sampled each year. For each site sampled in any given year, samples for nutrient water chemistry analysis were collected three to six times between June 15 and October 15, and the results expressed as the geometric mean for each measured parameter. Geometric means were used in lieu of arithmetic means given that nutrient concentrations had a log-normal distribution (i.e., an arithmetic mean taken from log transformed values yields a geometric mean in original units). Nutrients included in the analyses were nitrate-nitrite nitrogen (NOx-N), ammonia nitrogen (NH3-N), total phosphorus (TP), and total Kjeldahl nitrogen (TKN). The method detection limit for TP was 0.01 mg/l. Values below method detection limits (14% for TP, 61% for NH3-N) were halved. Other water chemistry parameters included total suspended solids. Laboratory methodology followed procedures in APHA (1992). Hourly DO concentrations were recorded for a 24 to 48 hour period at 86 sites with automatic data loggers (probe accuracies for DO are within +/- 0.3 mg/l). Data loggers were deployed a week prior to, or several days following a chlorophyll sample, to coincide with stable, low-flow conditions.

Benthic chlorophyll-*a* concentrations were measured from epilithic periphyton communities by scraping a known area (3.35 cm^2) from each of ten to twenty (usually fifteen) large gravel to cobble size rocks from a glide-riffle-run complex. Methodology followed that discussed in detail in Moulton and others (2002), Scrimgeour and Chambers (2000), Cattaneo et al. (1997), and Lohman and others (1992). Only rocks that were undisturbed, as determined by a distinct, bi-colored appearance between the exposed surface and the side facing the stream bed, were collected. Rocks were collected once per site from late July to early September at a minimum of 10 days following any significant rainfall to minimize effects from scouring (Biggs 2000, Lohman and others 1992). Large gravel (> 7.5 cm diameter) to cobble sized substrates were chosen to minimize potential spatial variation within the stream reach (Cattaneo and others 1997). The rock scrapings were combined and blended with a rechargeable Cuisinart® (East Windsor, NJ, USA) model CSB-77 hand blender. Three 5 ml aliquots were drawn from the slurry and each filtered on Whatman® GF/C 1.2 micron glass fiber filters in the field, and either placed on ice for daytrips or frozen on dry ice for overnight trips. The chlorophyll on the filters was extracted using a known quantity (10–15 ml) of 90 percent acetone. The amount of chlorophyll-*a* and pheophytin a in a sample was determined using EPA Method 445 (USEPA 1997). Calibration of the fluorometer was against a known standard. Results from each of the three filters were averaged, and the concentration of corrected benthic chlorophyll-*a* or pheophytin a (hereafter referred to as benthic chlorophyll and pheophytin, respectively) at a given site was expressed in mg/m^2 as extrapolated from the slurry volume and total rock area scraped. Precision of the method, as relative standard deviation (RSD) is given in Method 445.0 as typically between 25–30 percent. The estimated detection limit for the fluorometric method ranges from 50 picograms per ml (5x10-7 mg/l) as quoted by the manufacturer to 0.1 mg/l for a pooled estimated detection limit [USEPA 1997 (p-EDL)]. Due to the large dilutions required to analyze these solutions, the fluorometric p-EDLs are unrealistically high compared to what is achievable by a single lab. Typical single lab EDLs can easily be 1000-fold lower than the p-EDL reported here (USEPA 1997). The range of chlorophyll-*a* concentrations anticipated for the streams in this study is estimated at 10 to 300 mg/m^2; 10 mg/m^2 has a volumetric equivalent of 0.157 mg/l, assuming 15.7 cm2 of surface is scraped and placed in a one liter of water.

Stream physical habitat quality was assessed using the Qualitative Habitat Evaluation Index (QHEI; Rankin, 1995) at least once in a given year. The QHEI is a qualitative visual assessment of functional aspects of stream macrohabitats (e.g., amount and type of cover, riparian width, siltation, channel morphology). An estimate of light availability at a site was given by the degree of open arc between the canopy tops of either bank. A clinometer was used by an observer standing in the middle of the stream channel to measure the angle to the canopy top of opposite banks at three locations within the sampling reach. The sum of the two measured angles were subtracted from 180, and averaged for the three observation points to yield what is hereafter referred to as canopy cover.

Fish communities were sampled once at 100 of the 109 sites using generator powered, pulsed DC electrofishing units and a standardized methodology (Ohio EPA 1987, 1989a, 1989b; Yoder and Smith 1999). Samples were typically collected within 2 weeks following chlorophyll samples, under the same flow conditions. Fish community attributes were collectively expressed by the IBI (Karr 1981; Karr and others 1986), as modified for Ohio streams and rivers (Yoder and Smith 1999; Ohio EPA, 1989a). Macroinvertebrates were sampled quantitatively at fifty-six sites using modified, multiple-plate artificial substrate samplers (fashioned after Hester and Dendy 1962), and sampled qualitatively for presence/absence at 102 sites (that included the previous 56). The artificial substrates were deployed 2 to 3 weeks prior to, and 3 to 4 weeks post chlorophyll sampling. Qualitative samples were collected when the artificial substrate were retrieved (i.e., not all artificial substrates are retrieved, hence the disparity in sample types). Macroinvertebrate community structure for quantitative samples was expressed as the Invertebrate Community Index (ICI; DeShon 1995). The ICI is a multimetric measure of the invertebrate community composed of ten metrics scoring functional, compositional and taxonomic attributes. ICI scores were binned into eight ranks based on narrative ranges (e.g., excellent, very good, good, etc.). For the 46 samples with only presence/absence data, staff biologists assigned one of the eight ranks to the samples based on both the relative composition of macroinvertebrates in the sample, and weighted tolerance values for individual taxon (DeShon 1995). The weighted tolerance values for individual taxon were derived from weighted average ICI scores; thus, the narrative assignments correspond to ranges of ICI scores (DeShon 1995). Hereafter, the rankings are referred to as the invertebrate community (IC) ranks. The number of taxa in the orders Ephemeroptera, Plecoptera, and Trichoptera (EPT) from qualitative samples was also used as a measure of community quality.

2.1.3 Statistical Analyses

Values for benthic chlorophyll, pheophytin, water chemistry parameters, stream gradient, drainage area, percent urban land use, and canopy were log10 transformed to normalize distributions prior to statistical analyses. Percent agriculture was normally distributed. Relationships between benthic chlorophyll concentrations and the aforementioned variables were initially described by simple Pearson correlations and inspection of scatter plots. One site collected from the Mahoning River was identified as an outlier (by the Hadi [1994] algorithm of SYSTAT), and excluded from all subsequent analysis. Of the environmental variables identified as having a marginal ($P < 0.1$) or better ($P < 0.05$) association with benthic chlorophyll, all subsets regression (Neter and others 1990) was used to suggest linear combinations that explain variation in benthic chlorophyll given dissolved inorganic nitrogen (DIN, the sum of ammonia nitrogen and nitrate-nitrite nitrogen) and canopy cover. This exercise was not done to find a predictive equation; rather it was to inform change point analysis by identifying which environmental variables account for variation in benthic chlorophyll beyond that explained by nutrients and light. DIN and canopy were forced a priori given that nutrients and light are well-established predictors of benthic chlorophyll in small streams. DIN was used in lieu of NOx as the two are statistically equivalent (see Table 1), and DIN would likely be used in management. DIN was forced in favor of TP because it showed a stronger association with benthic chlorophyll (Table 1). However, residuals from the regression of TP on DIN were used as a free predictor to assess the explanatory contribution of TP uncompromised by multicollinearity (Graham 2003). The model with the lowest value for Mallow's Cp, the smallest number of predictor variables, and the highest adjusted coefficient of determination was subsequently run excluding data from the 19 large river sites to gauge changes in the explanatory power and slopes of the various parameter estimates, given that the 19 large river sites were potentially transitional between periphyton and phytoplankton dominated systems.

Change points in benthic chlorophyll concentrations in relation to TP or DIN were identified by first obtaining the residuals from the regression of benthic chlorophyll concentrations on canopy cover and percent agricultural land use, given that the latter was consistently identified as a predictor in all subsets regression. The residuals then served as a dependent variable in a regression tree where either TP or DIN was an independent variable. The trees were constrained to a single split and a minimum of 10 cases (i.e., ~ 10 percent of the sample) in a terminal node. A change point in benthic chlorophyll in relation to canopy cover was similarly obtained using the residuals from the regression of benthic chlorophyll on DIN and percent agricultural land. The reduction in variance afforded by the change point identified by each regression tree was gauged using an F-test (Qian and others 2003). Note that the F-tests were not formally testing respective null hypotheses of similar variances as the change points were not chosen beforehand.

An estimate of uncertainty in the cut point for each regression tree model was evaluated with a 1000 count bootstrap sample (Qian and others 2003). To help interpret results, frequency histograms of cut values from the bootstrap samples were overlain onto scatter plots of benthic chlorophyll residuals and each of the three independent variables. Also, for each scatter plot, a locally weighted line was fitted to the data using the LOWESS (q=0.5) function in SYSTAT (San Jose, California, USA).

Information from automated monitoring of DO at a sampling location was summarized as the maximum range in concentration, and the minimum value recorded over a 24 hour (h) period for a given location. Linear models explaining variation in DO range were suggested by all subsets regression that included benthic chlorophyll in all models, and pheophytin, stream gradient, and QHEI scores as free predictors. Pheophytin was introduced as an independent variable because it serves as the primary electron receptor in photosystem II (Marshall and others 2000), and is thus an important accessory pigment in the living fraction. Also, as a measure of the senesced fraction, pheophytin may represent the potential for oxygen demand, and thus serve as a proxy for daily swings in DO. However, because pheophytin was strongly correlated with benthic chlorophyll, residuals from the regression of pheophytin on benthic chlorophyll were used in lieu of the measured values (Graham 2003). Stream gradient was included as a rough proxy for re-aeration, and QHEI scores were included to account for variation due to overall physical habitat quality. Results from the all subsets regression indicated that pheophytin residuals and QHEI scores formed a parsimonious set of predictor variables; therefore, residuals from the regression of DO range on QHEI scores and pheophytin residuals were used in change point analysis. A change point in DO range over benthic chlorophyll was given by a regression tree constrained to a single split and a minimum of 9 cases (i.e., 10 percent of the sample) in a terminal node. Uncertainty was evaluated by overlaying a frequency histogram of cut values from a 1000 count bootstrap sample on a scatter plot of DO range (residuals) over benthic chlorophyll concentrations in concert with a LOWESS (q=0.5) fitted line. Minimum DO concentrations were assumed a priori to be largely a function of DO range, and therefore regressed against the DO range, stream gradient and drainage area.

Indicators of macroinvertebrate and fish community quality were regressed against indicators of nutrient enrichment, either benthic chlorophyll concentration, the range of daily DO concentration, or daily minimum DO concentration, to test whether a linear relationship existed between any of the biological and enrichment indicators. For the macroinvertebrate community, community rankings and the number of EPT taxa were used as indicators of quality, and for fish, IBI scores and the number of sensitive fish species were used. Because habitat quality is a known predictor of the fish and macroinvertebrate indicators, QHEI scores were included as an independent variable in each of the regressions to ascertain if the enrichment indicators explained significant additional variation in the biological indicators. Similarly, drainage area was included as an independent variable in regressions between the number of sensitive fish species and the enrichment indicators. If an enrichment indicator explained variation in a biological indicator, change points between the two were identified with regression trees that followed the methods previously described for DO and benthic chlorophyll wherein residuals following regression on QHEI scores were the dependent variable.

2.2 Abridged Results of Published Nutrient Study

Benthic chlorophyll-*a* levels were directly related to nutrient concentration (Figures 3a and 3b) and the amount of canopy cover shading a given sampling location (Figure 3c). A change point in benthic chlorophyll-*a* level was detected when dissolved inorganic nitrogen concentrations exceeded 0.435 mg/l, and when total phosphorus concentrations exceeded 0.038 mg/l (Figure 3a and 3b). Although a change point in both cases was detected, note that the relationships between benthic chlorophyll and nutrients were monotonic. Variation in 24 hour DO range was significantly and monotonically correlated with benthic chlorophyll-*a*, but a distinct threshold response was evident when benthic chlorophyll-*a* levels exceeded 182 mg/m^2, at which point 24 hour DO swings exceeding 7 mg/l became frequent (Figure 4a). Minimum daily DO concentrations were strongly correlated with 24 h DO range ($p<0.00001$), such that 24 hour swings in excess of 7 mg/l carry a significant risk of minimum DO values falling below WQS (Figure 4b).

Macroinvertebrate community index (ICI) scores and EPT taxa richness were positively correlated with minimum DO, and negatively correlated with 24 h DO range (Figures 5a-d). Both invertebrate community indicators were negatively correlated with benthic chlorophyll-*a* levels. Fish IBI scores had a negative association ($0.05 < p < 0.10$) with minimum DO concentrations when all sites were considered. If coldwater sites were excluded, IBI scores were significantly negatively correlated with minimum DO (Figure 6). Thresholds or change points suggested by regression trees of the raw data and bootstrapped samples shown in Figures 3-5 are summarized in Table 4.

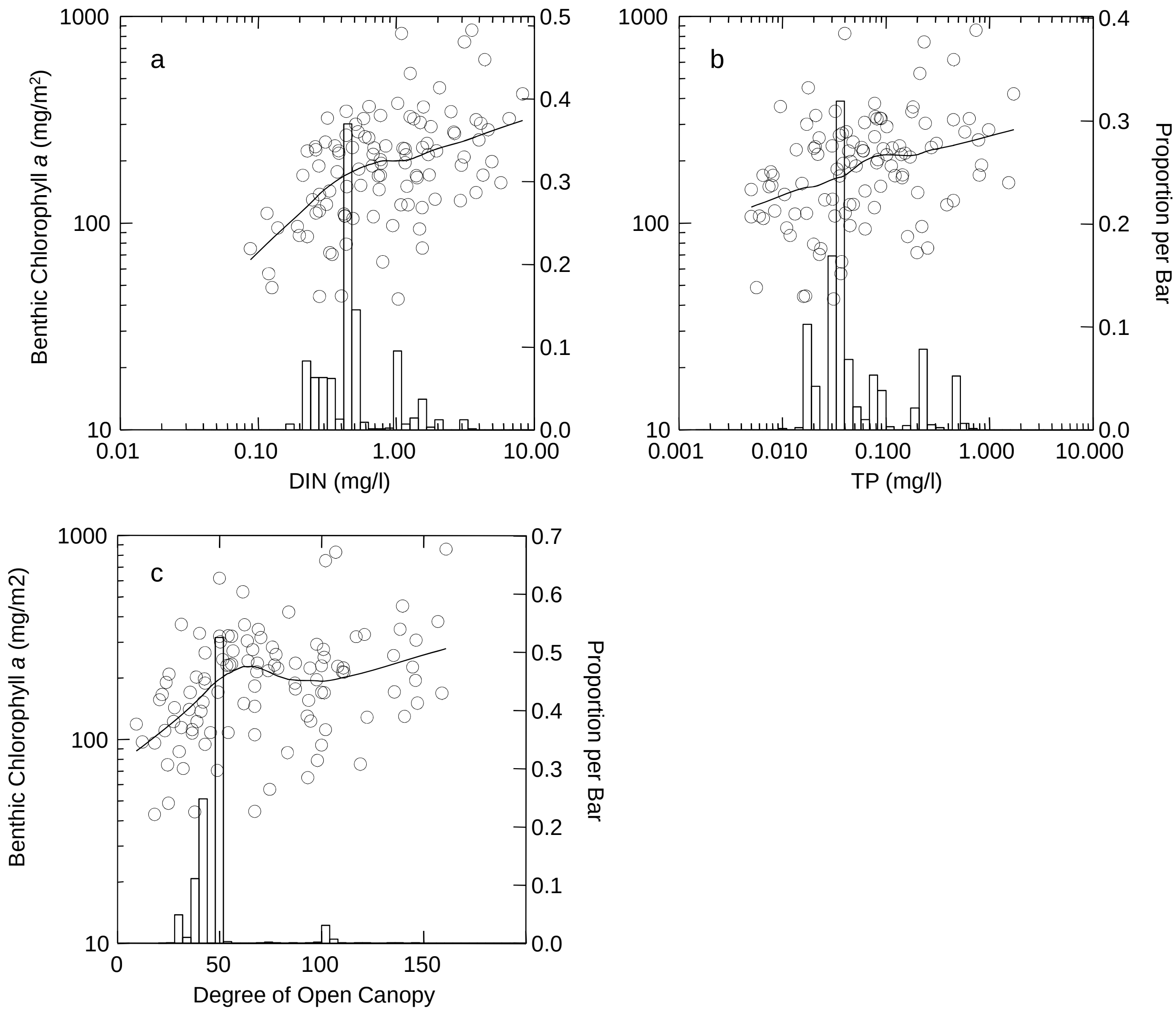

Figure 3. Benthic chlorophyll-*a* concentrations in relation to a) dissolved inorganic nitrogen, b) total phosphorus, and c) canopy cover. Lines following the local central tendency in each plot are from LOWESS (q=0.5), and the superimposed histograms show the frequency (right y-axes) distributions of regression tree cut values given from bootstrapped samples.

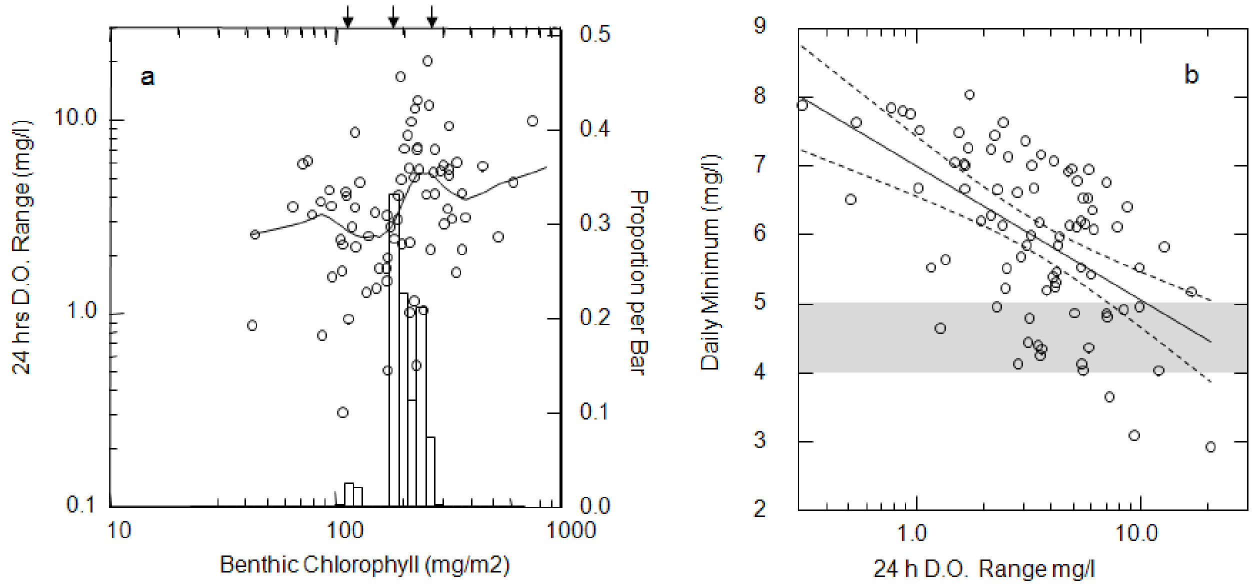

Figure 4. a) 24 hour range in DO concentrations in relation to benthic chlorophyll-*a* levels. For reference, arrows arrayed along the top of the graph demarcate the 25th, median and 75th percentile levels of benthic chlorophyll-*a*. The lines following the local central tendency is from LOWESS (q=0.5), and the superimposed histogram shows the frequency (right y-axis) distribution of regression tree cut values given from bootstrapped samples. b) Daily minimum DO concentrations plotted against 24 hour range in DO. The gray-shaded area shows the range of existing WQS for minimum DO. The solid line through the plot is from ordinary least squares regression, and the dashed line represent the 90% confidence interval of the regression line.

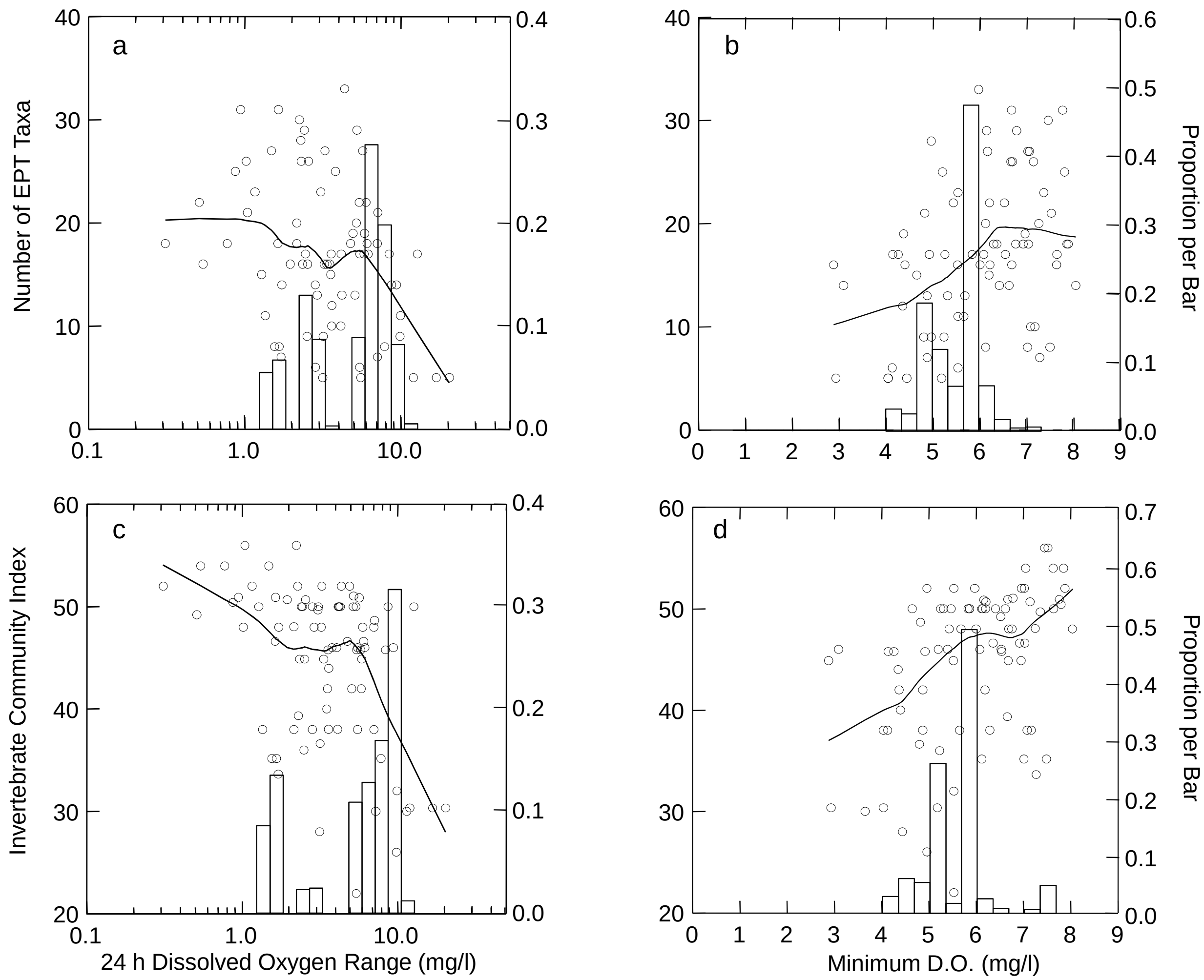

Figure 5. EPT taxa richness in richness in relation to a) 24 hour DO range, and b) minimum daily DO concentration; and ICI scores in relation to c) 24 hour DO range, and d) minimum daily DO concentration. Lines following the local central tendency in each plot are from LOWESS (q=0.5), and the superimposed histograms show the frequency (right y-axes) distributions of regression tree cut values given from bootstrapped samples.

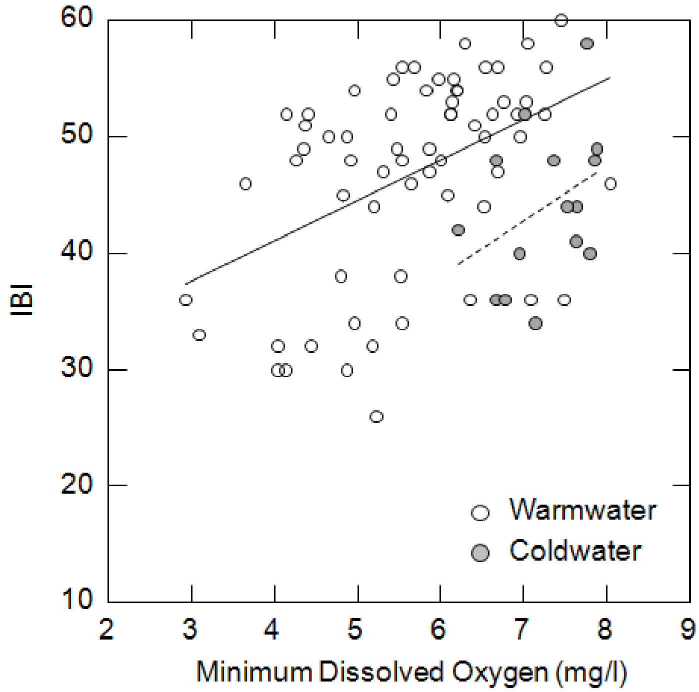

Figure 6. IBI scores in relation to minimum DO concentration. Regression lines are from a separate slopes model using a coding variable to indicate coldwater or warmwater streams. If the coding variable is introduced into a regression of IBI on QHEI scores and minimum DO concentration, all terms are significant ($p < 0.01$) and the model explains 35% of the variation in IBI scores.

Table 4. Estimates of uncertainty surrounding change points suggested by regression trees. The change point is the point in the X variable that divides the corresponding Y variable into two groups. Medians, seventy-fifth and ninetieth percentiles are from a 1000 count bootstrap sample. The F-test is for the difference between the variance in Y and the variance in Y when partitioned by the change point.

	All Data				Bootstrap Samples			
Y	X	N	Change Point	F	Median	Mode	75^{th}%	90^{th}%
Chl-a	DIN mg/l	108	0.435	11.125	0.435	0.435	1.095	1.556
Chl-a	TP mg/l	108	0.038	8.585	0.038	0.038	0.048	0.078
Chl-a	Canopy degrees	108	40.0	10.151	41.0	40.0	50.0	84.0
DO Range	Chl-a mg/m^2	85	182.0	6.874	194.0	190.0	196.0	231.0
EPT Richness	Chl-a mg/m^2	102	107.0	5.722	111.0	96.0	122.0	214.0
ICI	Chl-a mg/m^2	102	320.0	2.484	261.0	320.0	320.0	365.0
EPT Richness	Min. DO mg/l	83	5.86	5.459	5.86	5.86	5.86	6.14
ICI	Min. DO mg/l	83	5.25	4.534	5.31	5.20	5.86	7.52
EPT Richness	DO Range mg/l	83	7.04	3.347	2.87	7.04	7.04	7.85
ICI	DO Range mg/l	83	9.36	6.389	8.69	9.85	9.85	9.85

3. Supplemental Analyses

3.1 Structural Equation Modeling

The preceding results are an abridged version to show the basic casual pathway of enrichment from primary nutrients to the condition of biological assemblages, as the thresholds identified at individual steps along the path form the technical basis of water quality criteria for nutrients. It is important to understand the various factors that contribute nutrients to a system, and others that either facilitate or mute the expression of enrichment. Essentially, there are three components to consider. First, within the nutrient study data set, many of the variables are collinear (Table 5), and that complicates interpretation of results from traditional multiple regression (Graham 2003), especially when the end goal is to understand how the variables ultimately relate to biological condition. Second, in the case of nutrients, instantaneous measures from water quality grab samples are essentially point estimates of a larger, unmeasured phenomenon that might be described as a combination of antecedent loadings and ambient concentrations. Seasonal collections to estimate an average condition are a workaround, but not perfect. Similarly, although benthic chlorophyll-*a* is a direct measure of the expression of nutrient enrichment, it too can be thought of as an abstraction of a larger phenomenon. Here, chlorophyll-*a* is a proxy for periphytic algae, and algae influence DO through photosynthesis and respiration. However, other components of the microbial community, as well as physical factors influence DO regimes. So in this sense, chlorophyll-*a* (and pheophytin) represents an unmeasured (or latent) potential to influence DO. Third, and most obvious, are the individual and combined effects of the various measured parameters on the response variable of interest, namely, biological condition.

Structural equation modeling (SEM) is a technique that allows one to test and parameterize hypothesized relationships among measured and latent variables based on the variance/covariance structure of the data set (Graham 2003, Grace and Bollen 2005). In this regard, SEM is strictly a confirmatory technique; however, initial results can be used to redirect model elements toward a better fit in cases where the initial hypothesized model does not comport with the variance/covariance structure of the data set. SEM is similar to traditional multiple regression in that one of the objectives of analysis is to account for the variation in a response variable explained by a set of measured predictor variables. SEM, however, goes a step further by examining shared contributions of the predictor variables, rather than simply unique contributions, as is the case for multiple regression; an important distinction where multicollinearity exists among predictor variables. SEM can also be used to evaluate competing models, when those models are specified a priori.

An SEM was developed using AMOS 20 software for the nutrient data set based on the initial hypothesized relationships shown in Figure 7a. Estimation was by maximum likelihood, regression weights and covariances were evaluated for significance at the $p<0.05$ level using a 200 sample Monte Carlo bootstrap to obtain 90th percentile confidence intervals of respective estimates. The initial model was drawn based on a combination of observed and hypothesized relationships in the nutrient data set, and frequently observed associations between biological indicators and key environmental variables (e.g., ammonia-nitrogen, urban land use) or known relationships (e.g., EPT taxa richness and drainage area). The nutrient-specific component of the model consisted of the path from nutrients through the expression of enrichment to EPT taxa richness. As alluded to earlier, DIN and TP were considered observed measures of the temporally proximate aspect of nutrient loads. Percent urban and agricultural land uses were considered observed measures of the more temporally distal aspect of nutrient loadings. Benthic chlorophyll-*a* was considered a measured aspect of nutrient loads and related to the amount of canopy cover. Oxygen stress was a latent variable intended to capture the measured influence of photosynthesis, as given by 24 h DO range, and respiration or oxygen demand, as given by pheophytin, where pheophytin stands as a proxy for senesced algae. Pheophytin was related back to chlorophyll-*a*. DO stress was included as a latent variable because the direct measures of DO were point-in-time estimates, albeit over 24 hours, and because 24 h DO regimes are a function of multiple parameters including photosynthesis, respiration, reaeration, temperature, and surface and groundwater inputs. Minimum DO was considered an observed measure of DO stress. EPT taxa richness was related to minimum DO, QHEI, ammonia-nitrogen (general association), percent urban land use (general association, Yoder et al. 2000), and drainage area (DeShon 1995).

An initial run of the data against the model resulted in inconsistencies between the model and the variance/covariance structure of the data (Figure 7b). Modification indices suggested (after several iterations) the final structural model shown in Figure 8. Model fit indices (Table 5) suggested that the final model is consistent with the variance/covariance matrix. The final model also generally comports with the initial model in overall tone, but differs in several important details. Most strikingly, percent agricultural land use is removed as an observed variable of Nutrient Load, and redirected to having a direct relationship with DO Stress. Also, benthic chlorophyll-*a* is now not a direct predictor of 24 h DO range, but mediates DO Stress through pheophytin, and has an indirect effect on EPT taxa richness through pheophytin. Examination of the bivariate relationships among percent agricultural land use, 24 h DO range, canopy cover, and benthic chlorophyll-*a* (Figure 9) shows no association between canopy cover and percent agricultural land use, but a strong association between 24 h DO range and agriculture. Apparently, localized, well-canopied reaches can be light limited, but still experience wide DO swings because of upstream conditions. As a further illustration of the point, an aerial photograph of a typical stream (North Fork Massie Creek) in the Eastern Cornbelt Plain (Figure 10) shows how canopy (and channelization) can change abruptly across property or field lines. Percent agricultural land use may also serve in some capacity as a proxy for one or more of the parameters known to affect the 24 h DO regime. For example, tile drainage alters surface and groundwater inputs, land application of manure may influence respiration, and channelization my affect reaeration. Note, however, that the relationship between agriculture and nutrient load was indirectly retained through total phosphorus.

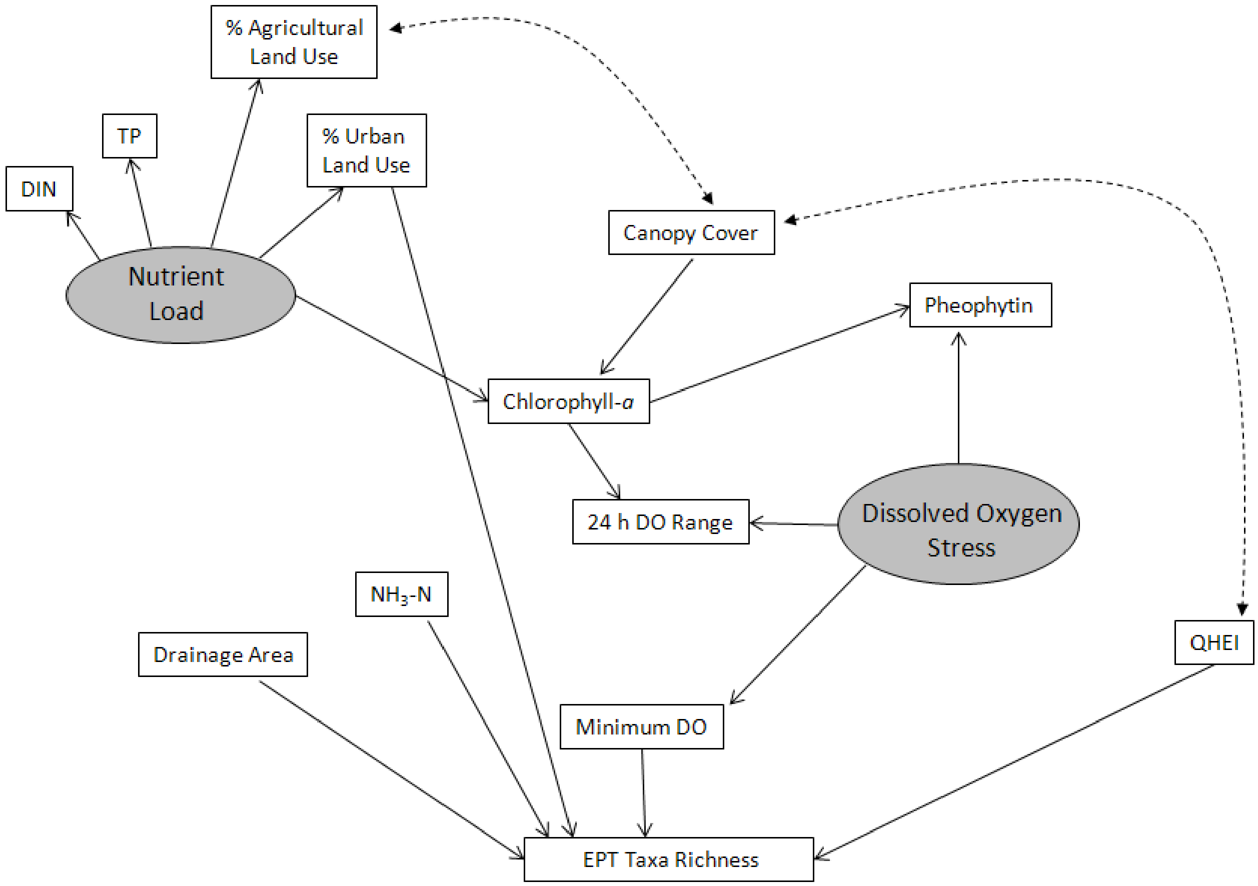

Figure 7a. The initial measurement model used to test relationships among variables in the nutrient data set. Variables placed in rectangular boxes are observed variables (i.e., those that were measured), and those placed in shaded ovals are latent variables. Solid arrow-tipped lines drawn between variables represent direct relationships. Stippled lines indicate variables with correlated error terms (i.e., correlated residuals).

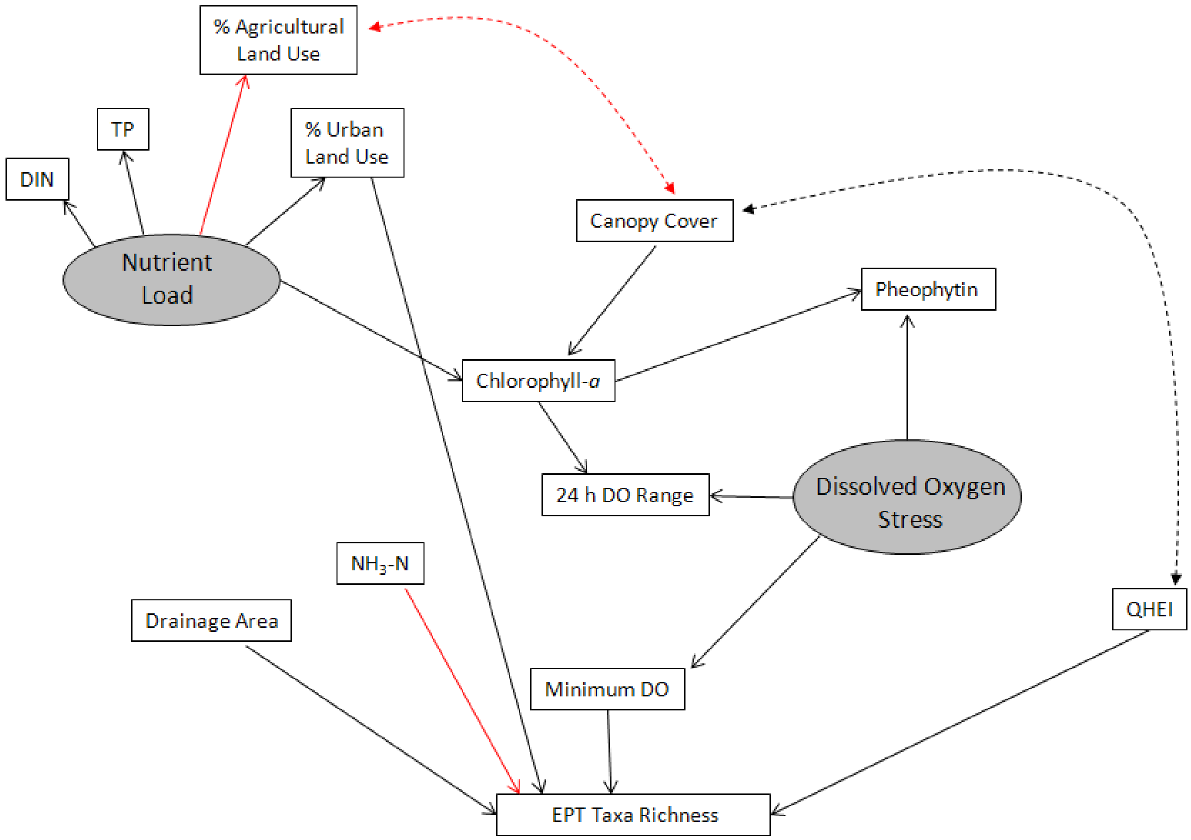

Figure 7b. The initial measurement model with hypothesized paths shown to be inconsistent with the variance/covariance structure of the data based on SEM.

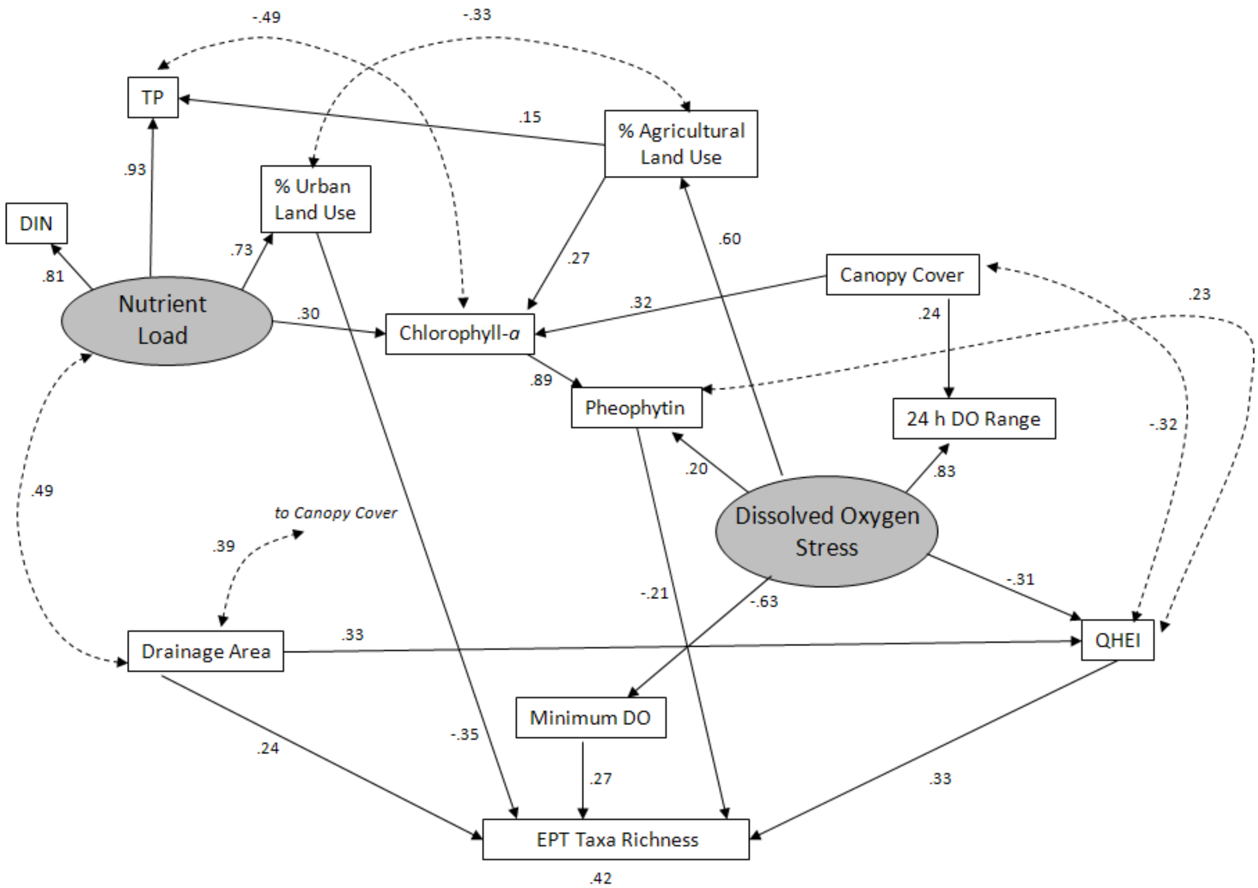

Figure 8. Final SEM for the nutrient data set where EPT taxa richness is the modeled biological response variable. Numbers adjacent arrows are standardized coefficients[2]. The model accounted for 42% of the variance in EPT taxa richness.

Table 5. Model evaluation indices.

Mode Fit Index	Value for Final SEM	Criterion for Good Fit
χ^2	χ^2=30.72, 39 df, p=0.83	p > 0.05
RMSE	0.00 (0.00-0.05, 90% CI)	RMSEA < 0.05
CFI	1.0	CFI ~ 1.0

[2] A standardized coefficient represents the change, in units of standard deviation, one variable has on another. For example, as percent urban land use increases by one standard deviation, EPT taxa richness is expected to decrease (notice the negative sign) by 0.35 standard deviations.

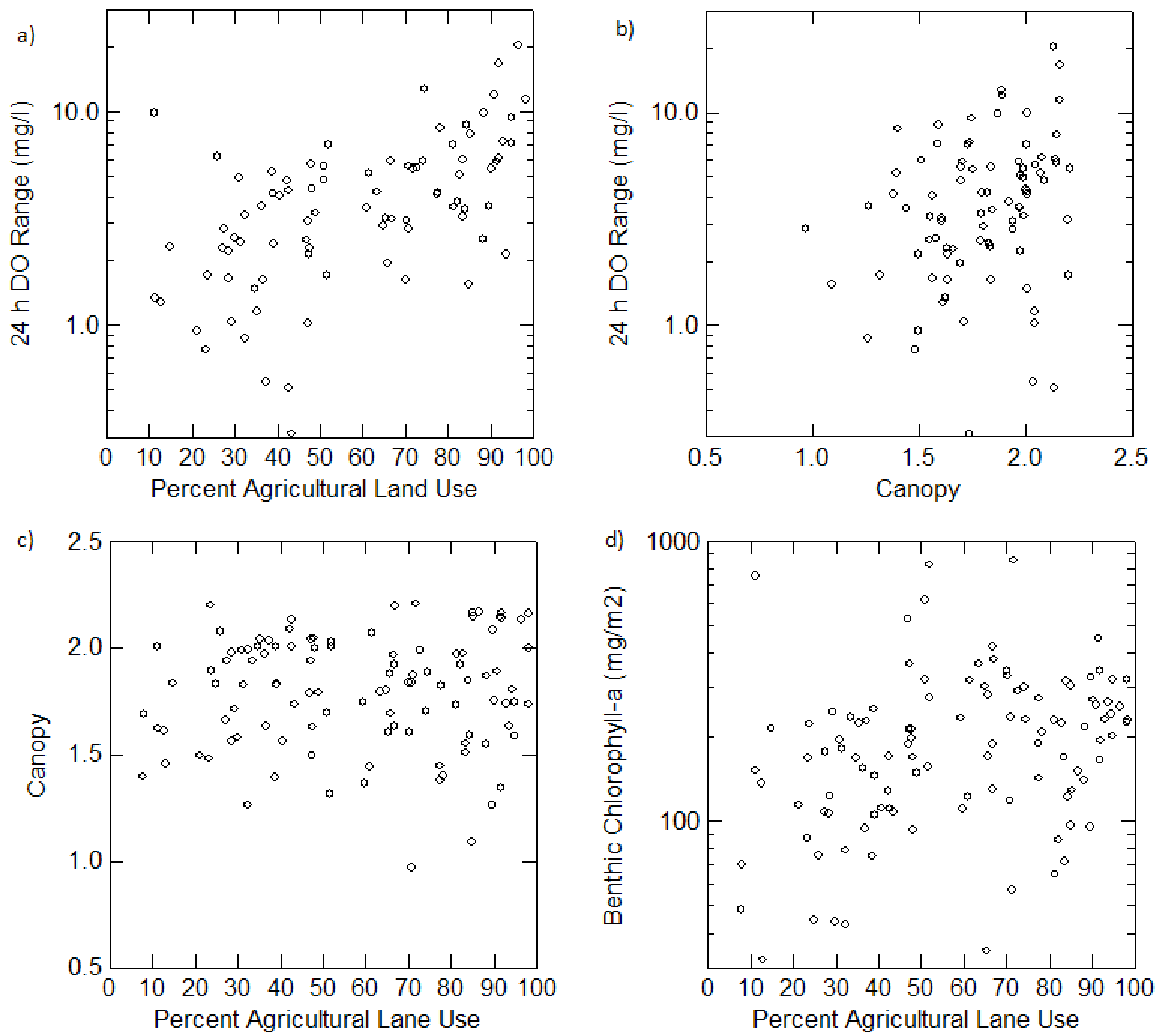

Figure 9. Bivariate plots of a) 24 h DO range on agriculture, b) 24 h DO range on canopy, c) canopy on agriculture, and d) benthic chlorophyll on agriculture.

Figure 10. Aerial photograph of the North Fork Massie Creek (83° 45' 7"W, 39° 46' 21"N). The historic meander belt and adjacent, now-drained wetlands are clearly demarcated by dark-colored soil. Recent channelization is evident through the property where the stream enters the picture on the right side of the photograph. Canopy cover begins abruptly at the property line near the center of the picture.

Another difference between the initial measurement model and the final structural model is that habitat quality is, in a manner of speaking, a measured component of DO Stress, sharing an inverse relationship. Lastly, total phosphorus and chlorophyll-*a* have correlated error terms. This is a manifestation of the tendency for phosphorus to be depleted by the periphytic mat. Evidence for this can be seen in plot of benthic chlorophyll-*a*, TP, and DIN:TP ratio against DIN (Figure 11). Benthic chlorophyll increases across the range of DIN values, as does TP. However, locally, the relationship between TP and DIN is flat up to ~ 0.7 mg/l DIN, and some of the lowest measured TP values corresponded to relatively high levels of benthic chlorophyll, and modest levels of DIN, but the highest DIN:TP ratios. As can be inferred from the relationships evident in Figure 11, nitrogen and phosphorus generally appear to co-limit benthic algal biomass.

Further evidence for, and the implications of, co-limitation can be seen when benthic chlorophyll-*a* concentrations are stratified by DIN:TP ratios <>15 and plotted against TP or DIN (Figure 12). Relative to TP, benthic chlorophyll-*a* levels increase across the range of TP concentrations, but are higher per unit level of phosphorus at P-limited sites relative to N-limited sites. In other words, when nitrogen is relatively scarce, chlorophyll levels tend be lower across the range of phosphorus concentrations , but still vary with TP concentrations—evidence for co-limitation. Relative to DIN, chlorophyll levels also increase across the range of concentrations, but no difference is evident between N- and P-limited sites. The upshot of this is that it even for N-limited sites, reducing phosphorus should have a measurable effect. This is an important implication given the difference in costs of P-removal compared to N-removal.

The final measurement model shown in Figure 8 resulted from following suggested modification indices. Redirection of paths, or specifying new paths within a model, should only be done if there is a plausible mechanistic reason. However, given the multitude of factors influencing any particular reach of stream, several or more plausible explanations can be had for any number of suggested paths. So although none of the paths shown in Figure 8 may necessarily be implausible, some of the suggested modifications may have been due to chance relationships in the data, and a fanciful rendering of plausibility, resulting in overfitting of the model, especially with respect to the latent variable DO Stress. Again, however, the final model comports with the initial model in overall tone. By following the modification indices, a potential weakness (though not, emphatically, a detraction) was revealed in lacking measures of reaeration and respiration. Conversely, depletion of TP by periphyton was revealed, as was a more direct relationship between periphyton biomass and EPT taxa richness. In this context, the consequences of overfitting are negligible, but serve to expose gaps in an overall mechanistic understanding. Note that overfitting does not alter the standardized regression coefficients leading to EPT taxa richness, nor the overall explanatory power of the model.

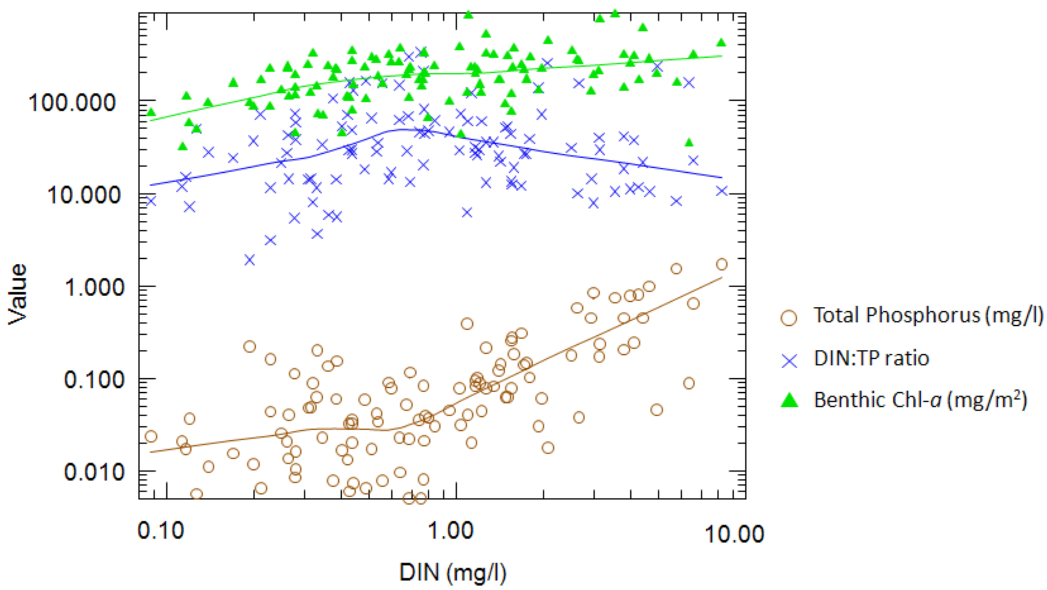

Figure 11. Total phosphorus concentrations, DIN:TP ratios, and benthic chlorophyll-*a* levels plotted against dissolved inorganic nitrogen concentrations. Lines following the local central tendencies are drawn from LOWESS (q=0.5).

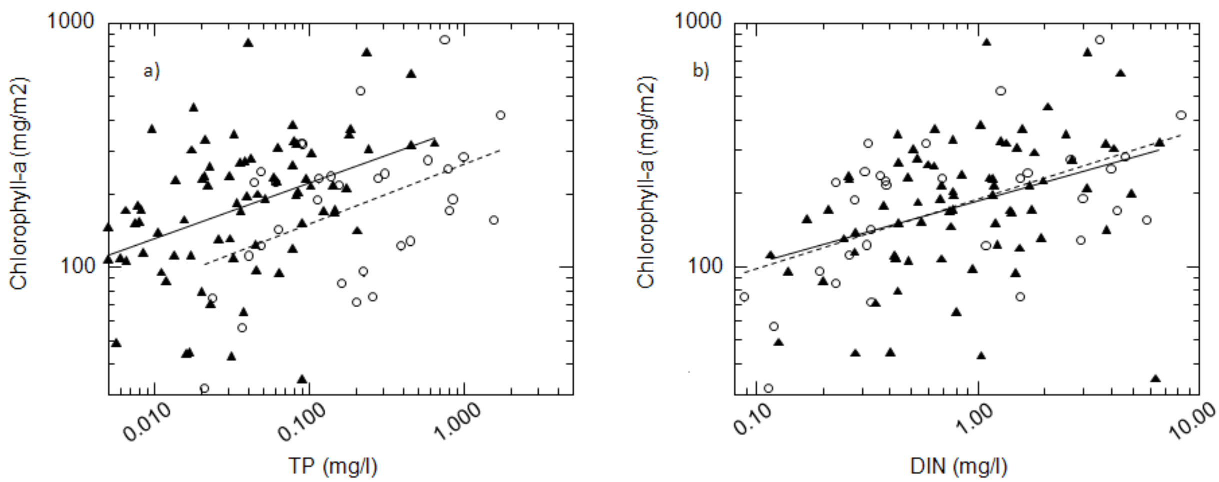

Figure 12. Benthic chlorophyll plotted against a) total phosphorus and b) dissolved inorganic nitrogen as a function of N:P ratios. The solid lines and filled triangles show P-limited (N:P >15) sites , the open circles and dashed lines show N-limited sites. Fitted lines are from linear regression. The lines in a) differ with respect to the Y-intercept ($p < 0.02$) from a linear regression of benthic chl-a on TP using an indicator variable (0,1) to denote N:P ratios < or > 15. This result is similar to what Dodds et al. (2002) found generally for temperate streams. The slopes and intercepts in b) obviously do not differ.

3.2 Logistic Regression

The nutrient study was designed to trace the causal pathway from primary nutrients through intermediaries to the effect on biological communities, and to demarcate diagnostic thresholds or benchmarks along the enrichment continuum. Ultimately, however, when the continuum is abstracted to a single indicator given by phosphorus or nitrogen, the point along the continuum where designated beneficial uses are likely to become impaired is a necessary component of water quality criteria for setting discharge limits and calculating load allocations. We examined Ohio's historic database to determine whether relationships identified in the nutrient study held true in the historic data, given that several variables used in the nutrient study analysis had not been routinely monitored prior to the nutrient study. The analysis focused on associations between measured nutrient concentrations (DIN and TP) and the biological endpoints of fish IBI and macroinvertebrate ICI. Periphyton chlorophyll *a*, minimum DO, and DO range were not measured in the historic data, and could not be part of the analysis.

Logistic regression was used to estimate the probability of fish and macroinvertebrate biotic indices meeting respective biological criterion given TP or DIN as stressors. Logistic regression is a linear method that tests explicit outcomes in one variable in relation to another, where the explicit outcomes are essentially either-or statements. Because Ohio has defined numeric standards for biological measures, outcomes of biological response against a stressor can be defined as the biological measure meeting the standard, or not meeting the standard, and expresses as a binary variable (i.e., 1 for meeting, 0 for not meeting). The proportion of ones to zeros along the ordered stressor variable are used to estimate probabilities of meeting or not meeting the biocriterion as the stressor variable increases from low to high values.

Data used in the analyses were from Ohio EPA's biological and water quality databases, spanning the years 1982 through 2010. Data were screened to exclude polluted sites (ammonia-nitrogen >0.1 mg/l) and to remove outliers. Two basic models for each biological index were constructed, one testing the simple bivariate relationship against each nutrient stressor, and the other introducing habitat quality (QHEI scores) as an additional predictor variable. For the first model, data were selected for QHEI scores > 60 to minimize habitat as a covariate. For the latter model, all QHEI scores were included. Habitat quality has an obvious association with biological index scores, and as such, is a necessary dimension to consider when examining what amounts to an essentially *in situ* relationship between a biological assemblage and a single chemical gradient. Similarly, a set of logistic models were estimated using EPT taxa richness as a response variable, where the binary response was defined as ≤ or > 10 taxa. Insects in the orders Ephemeroptera, Plecoptera and Trichoptera (EPT) tend to be highly sensitive to disturbance and chemical stressors, thus representing a response variable potentially sensitive to the lower range of the nutrient gradient.

Results from the logistic regression models are summarized in Table 6, and shown graphically in Figures 13, 14 and 15. With respect to the biotic indices, the probability of meeting the Warmwater Habitat (WWH) biocriteria varies with respect to the QHEI. Relative to the mean QHEI score in the extant data (QHEI = 64), respective TP concentrations corresponding to a 0.5 probability of meeting WWH were lower for the fish IBI, and higher for the macroinvertebrate ICI. For the simple bivariate models, the TP and DIN concentrations corresponding to the 0.5 probability of meeting WWH were similar for the fish IBI and macroinvertebrate ICI. With respect to DIN, and to a lesser extent TP, the macroinvertebrates show a subsidy-stress response, where the probability of meeting WWH increases with increasing DIN concentrations, presumably due to a stimulatory effect, before a threshold is reached and a stress response is induced. The logistic models including TP were more robust than those including DIN, though neither had very good predictive ability[3]. The logistic models for EPT taxa richness were suspect as the thresholds occurred close to the edge of the data range, though the subsidy-stress response was evident in the EPT richness model for DIN as well (Figure 15). In lieu of logistic regression, quantile regression plots (Figure 15) of the residuals from a regression of EPT taxa richness on QHEI were examined to determine the point where taxa richness becomes less than expected relative to TP and DIN. Collectively, these results suggest that for waterbodies possessing good to excellent habitat quality (QHEI > 64), TP concentrations < 0.3 mg/l and DIN concentrations < 3.0 mg/l should maintain the WWH use. Where the use is EWH, TP concentrations less than 0.06 mg/l would be protective. For waterbodies where the habitat is marginal or compromised, TP concentrations < 0.13 mg/l are needed to maintain the basic aquatic life use.

Table 6. Concentrations of total phosphorus (TP) and dissolved inorganic nitrogen (DIN) corresponding to a 0.5 probability of biological indicators meeting predefined outcomes (WWH for the IBI and ICI).

	Fish IBI		Macroinvertebrate ICI		EPT Taxa Richness		
	QHEI=64	**QHEI>60**	**QHEI=64**	**QHEI>60**	**QHEI=64**	**QHEI>60**	**Quantile**
TP	0.131	0.400	0.207	0.358	ns	ns	0.06
DIN	3.62	6.7	6.75	10	ns	ns	3.55

[3] See Appendix A for diagnostic statistics of model output. Note that predictive ability refers to the ability of a given model to predict an outcome for a given set of observations. That does not obviate the central tendency of the model (i.e., the point where WWH is likely to become impaired given the extant data).

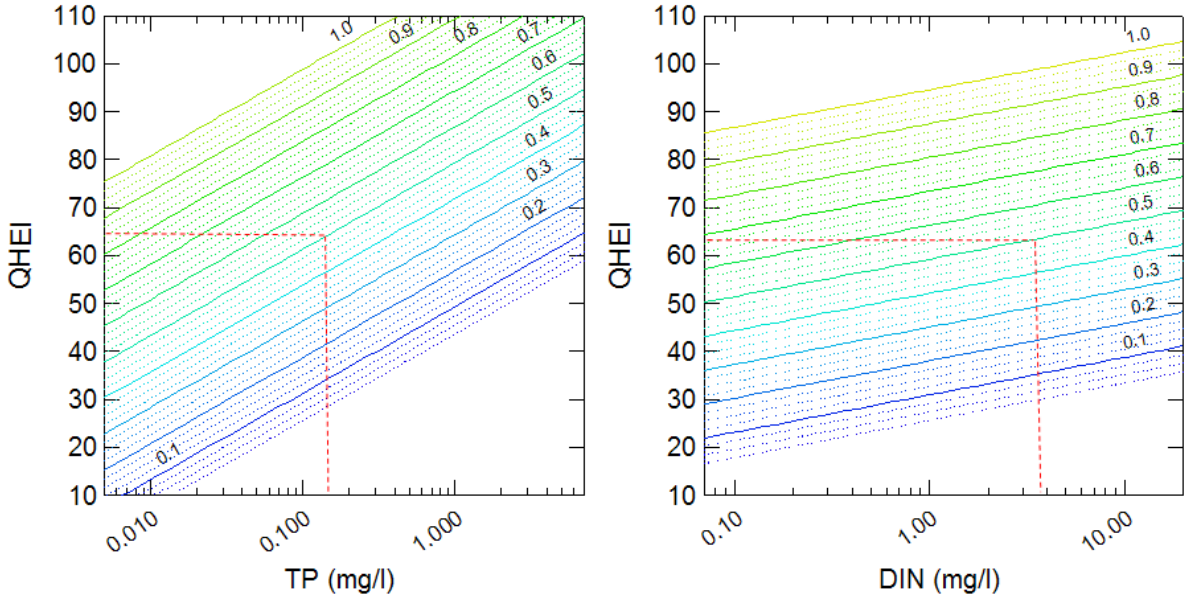

Figure 13. Results of logistic regression models for the fish IBI meeting WWH given total phosphorus and habitat quality (left panel), and dissolved inorganic nitrogen and habitat quality (right panel). Vertical lines drawn down to the x-axis correspond to the 0.5 probability of meeting WWH at the mean QHEI score of 64. The isolines show probability levels for a given combination of QHEI and TP or DIN values.

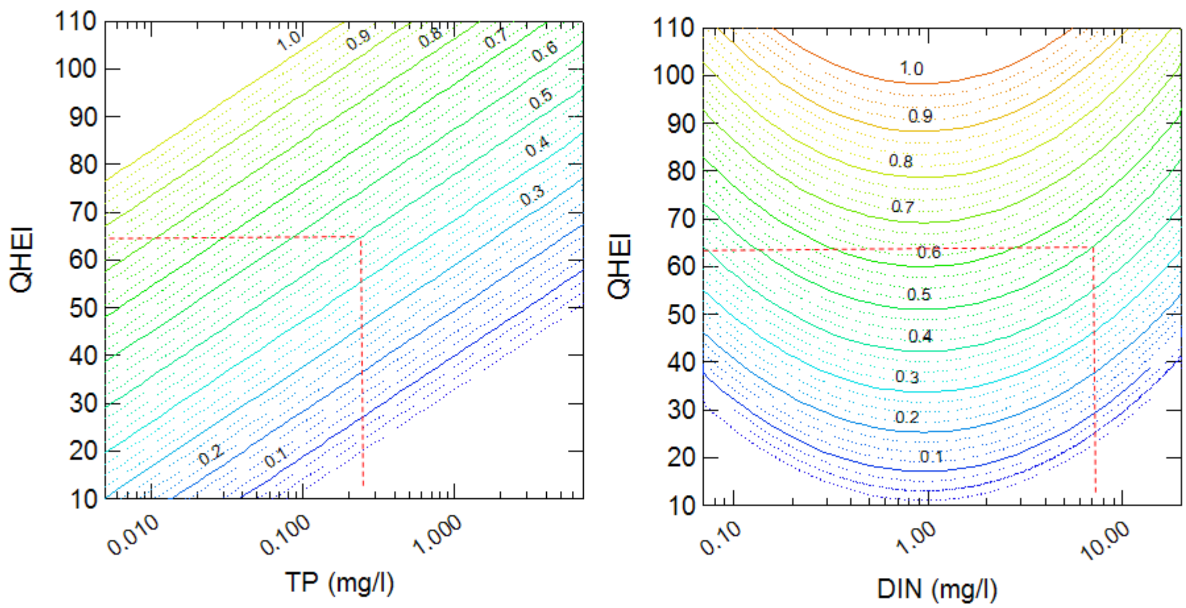

Figure 14. Results of logistic regression models for the macroinvertebrate ICI meeting WWH given total phosphorus and habitat quality (left panel), and dissolved inorganic nitrogen and habitat quality (right panel). Vertical lines drawn down to the x-axis correspond to the 0.5 probability of meeting WWH at the mean QHEI score of 64. The isolines show probability levels for a given combination of QHEI and TP or DIN values.

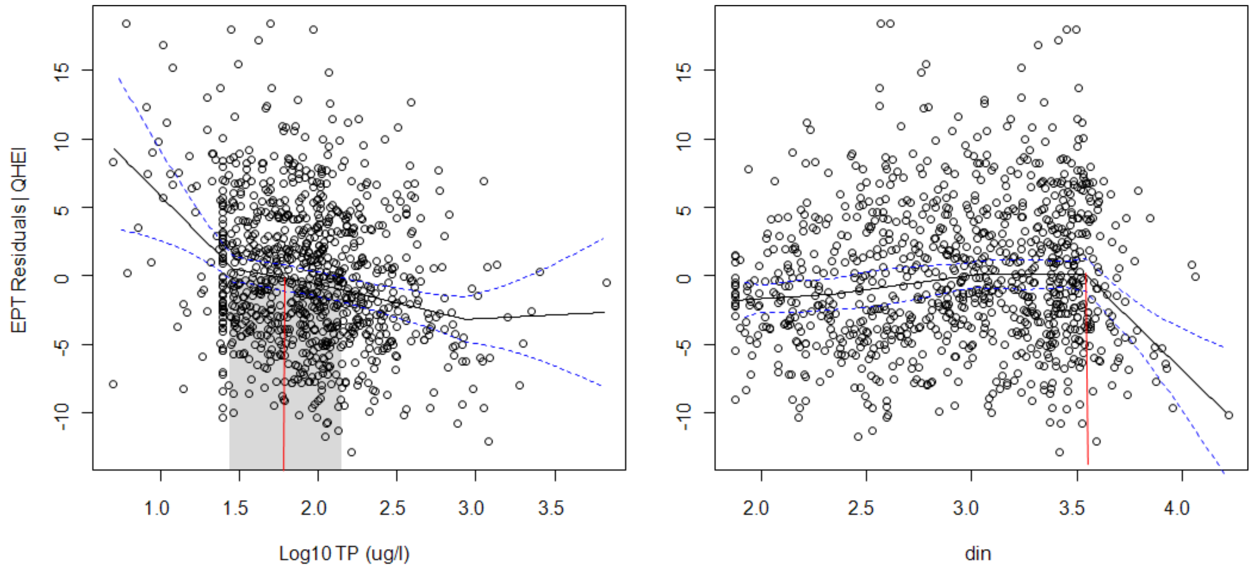

Figure 15. Quantile (tau=0.5) regression plots for EPT taxa richness residuals (following regression on QHEI) on total phosphorus (left panel) and dissolved inorganic nitrogen (right panel). The points where taxa richness is less than expected are noted by red lines drawn to respective x-axes. The shaded region in the TP plot shows where the 90th percent confidence bands cross the 0 point on the y-axis.

4. Synthesis of Field Study Results and Supplemental Analyses: The Trophic Index Criterion (TIC)

The field study identified benchmarks or thresholds leading from nutrients to biological condition via benthic chlorophyll-*a* (as a proxy for algal biomass) and DO regimes. The SEM model helps place those results in context against a wider representation of environmental variables by showing the inter-relationships among variables, and the relative strength of the various pathways. The results from logistic regression demarcate the TP or DIN concentrations where cumulative stress from enrichment has a better than even chance of resulting in biological impairment. These benchmarks, summarized in Table 7, now can be used to define where a given water body is positioned along a continuum of enrichment from background condition to over-enriched and impaired.

4.1 The Trophic Index Criterion

The Trophic Index Criterion (TIC) converts the thresholds and relationships of Table 7 into a numeric index by awarding points to the thresholds defined for each indicator such that, within a given indicator, levels typical of background conditions receive high scores, with successively lower points awarded as levels increase toward demonstrated impairment. Hence, the TIC provides a structured method of aggregating data collected on Ohio's streams and rivers into a numeric value that is essentially a translator for the condition of a waterbody relative to nutrient enrichment. As such, it can be used to dictate the imposition of appropriate nutrient management programs including NPDES permit limits, wasteload allocations, and abatement strategies for landscape pollution. Tables 7–10 list the metric scoring criteria for each TIC component metric. Because biological condition is the indicator most proximate to designated beneficial uses, it is weighed heavier in the TIC compared to the other components. Similarly, because the DO regime is both a more proximate stressor to biological communities, and a direct manifestation of nutrient enrichment, it is given a higher weighting than benthic chlorophyll-*a* and nutrients.

How the metric scores for nutrients and benthic chlorophyll-*a* relate to each other in light of the results from the SEM model are shown graphically in Figure 16. The SEM model suggests that as nutrient load increases by 1 standard deviation, benthic chlorophyll-*a* increases by 0.3 standard deviations. Similarly, the model suggests that as canopy cover (i.e., the degree of openness) increases by 1 standard deviation, benthic chlorophyll increases by 0.32 standard deviations. In Figure 16a, the TIC scoring ranges are superimposed on a scatter plot of TP on DIN concentrations, along with isopleths drawn according to standard deviations[4]. A nutrient metric score of 4 generally corresponds to -1 standard deviation, whereas a metric score of 1 or 0 is awarded when concentrations approach or exceed +1 standard deviation. In Figure 16b, isolines show how benthic chlorophyll-*a* levels vary over deviations in canopy cover and nutrient concentrations. The plot suggests that TP and DIN concentrations of less than 0.1 and 1.0 mg/l are required to prevent benthic chlorophyll-*a* levels from exceeding 182 mg/l in open canopied streams.

Table 7. Change points and thresholds between listed pairs of variables.

Stressor or Causal factor	Response				
	Benthic Chl-*a*	24 h DO Range	Minimum DO	Invertebrates	Fish
TP	0.04[a]			0.06[e]-0.65[c]	0.13-0.4[c]
DIN	0.44[a]			3.6-10.0[c]	3.6-6.7[c]
Benthic Chl-a		182 [a]		107 [a], 320 [a]	
24 h DO Range			7[b]	7.0 – 9.0 [a]	
Minimum DO				5.9 – 5.3 [a]	5.3[d]

a-change points defined by the nutrient study (Miltner 2010) and listed in Table 2
b-from linear regression (Miltner 2010), protective of existing WQS for minimum DO
c-ranges from logistic regression results appearing in Table 4.
d-change point based in IBI against minimum DO when coldwater sites are excluded
e-inferred from quantile regression

Table 8. Trophic Index Criterion scores for primary nutrients.

DIN (mg/l)	TP (mg/l)			
	≤ 0.040	0.041 ≤ 0.100	0.101 ≤ 0.300	> 0.300
≤ 0.44	4	2	1	1
0.44 < 1.01	2	2	1	0
1.01 ≤ 3.00	1	1	1	0
> 3.00	1	0	0	0

Table 9. Trophic Index Criterion scores for benthic chlorophyll-*a*.

Benthic Chlorophyll-*a* (mg/m^2)	Metric Score
≤ 107	4
107 ≤ 182	2
183 ≤ 320	1
> 320	0

[4] The standard deviation for nutrients was calculated based on total moles (DIN + TP) to normalize the concentrations for the isopleths in Figure 13a, and to have a common frame of reference for the x-axis in Figure 13b.

Table 10. Trophic Index Criterion scores for DO conditions.

Representative Dissolved Oxygen Concentration and Range (mg/l)	Metric Score
Minimum concentration is 5.0 or greater; 24-hour average concentration is 6.0 or greater; and 24-hour range is less than 6.0.	5
Concentrations meet criteria applicable to the use designation of the stream; and 24-hour range is less than 7.0.	3
Concentrations meet criteria applicable to the use designation of the stream; and 24-hour range is 7.0 to 9.0.	1
Concentrations do not meet criteria applicable to the use designation of the stream; or 24-hour range is greater than 9.0.	0

Table 11. Trophic Index Criterion Scores for biological condition.

Biological Condition	Score
All indices meet the exceptional warmwater habitat biological criteria.	6
All indices meet the warmwater habitat biological criteria; or The stream meets the definition of coldwater habitat.	4
The stream is designated limited resource water or limited warmwater habitat; or The stream is designated modified warmwater habitat and all indices meet the modified warmwater habitat biological criteria; or The stream is not designated limited resource water, limited warmwater habitat or modified warmwater habitat and all indices meet, or are in the nonsignificant departure range of, the warmwater habitat biological criteria.	1
The stream is designated modified warmwater habitat and one or more indices are below the modified warmwater habitat biological criteria; or The stream is not designated limited resource water, limited warmwater habitat or modified warmwater habitat and one or more indices are below the nonsignificant departure range of the warmwater habitat biological criteria.	0

Figure 17 is essentially a recapitulation of Figures 4 and 5 in light of TIC metric scoring, showing the relationships between benthic chlorophyll-*a* levels and 24 h DO range (Figure 17a), the DO regime in light of the SEM model (Figure 17b), and EPT taxa richness in relation to DO regime (Figures 17c and 14d). The distributions of 24 h DO in relation to benthic chlorophyll-*a* levels clearly shows the threshold at 183 mg/l (Figure 17a). The range in 24 h DO rarely exceeds 6 mg/l when benthic chlorophyll-*a* levels are less than 183 mg/m^2, and corresponding TIC metric scores for benthic chlorophyll-*a* would be $\geq$2 , and likely $\geq$3 for DO regime. Note, from Figure 17a, however, that high levels of benthic chlorophyll-*a* do not always correspond with wide DO swings, as over half of the measured ranges were less than 6 mg/l when the chl-a level was greater than 183 mg/m^2. Those cases show the advantage of a multi-component index, in that the chl-a metric would score ≤1, thereby down-weighting the final TIC score.

Figure 17b shows DO regime as function of standard deviations in the latent variable DO Stress from the SEM model. As DO Stress deviates from the mean by +1 standard deviation, conditions for a favorable DO regime are increasing unlikely—minimum DO concentrations begin to fall below established WQS, and 24 h DO range begins to exceed 6 mg/l. Note that if the minimum DO concentration falls below the WQS, the DO regime TIC metric score is 0. Obviously DO Stress is not a manageable entity, but the SEM model shows how the DO regime is influenced directly or indirectly by variables that are manageable, notably habitat quality and canopy directly, and nutrients indirectly. Lastly, Figure 17c and 14d show how favorable DO regimes correspond to biological condition as given by EPT taxa richness. A DO regime where the minimum DO is greater than the WQS and 24 h DO range is less than 6 mg/l (i.e., a TIC metric score of 5) typically supports biological assemblages consistent with beneficial aquatic life uses (i.e., biological condition TIC metric scores ≥4).

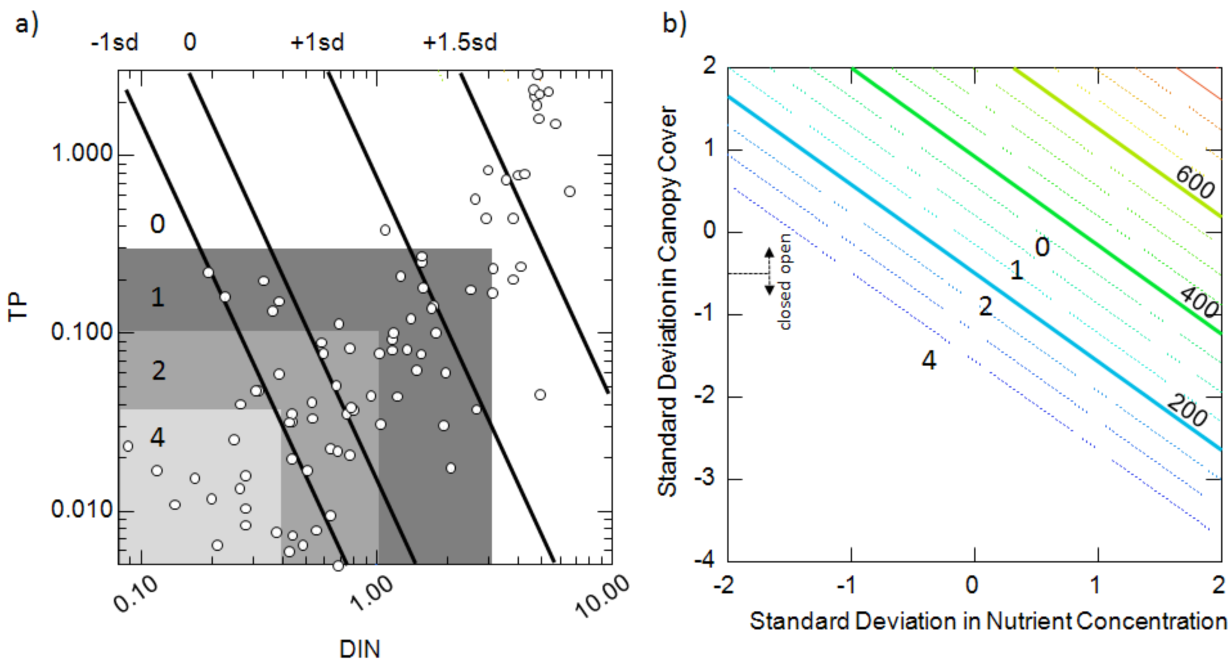

Figure 16. a) Scatter plot of total phosphorus concentrations on dissolved inorganic nitrogen concentrations (concentrations in mg/l) in relation to TIC metric scores and standard deviation in total molar nutrient concentration. The standard deviation for nutrients was calculated based on total moles (DIN + TP) to normalize the concentrations for the isopleths (a), and to have a common frame of reference for the x-axis (b). The 0 standard deviation line approximates the mean total molar concentration for a given combination of DIN and TP (i.e., in terms of total moles, the mean can stay constant as TP and DIN vary inversely). b) Benthic chlorophyll levels drawn as isolines in relation to deviations in nutrient concentrations (i.e., based on total moles), and canopy cover. TIC metric scores for benthic chlorophyll-*a* are superimposed on the graph. The threshold level of canopy cover where light becomes limiting (i.e., ~ 49^o arc in open canopy) is noted on the y-axis.

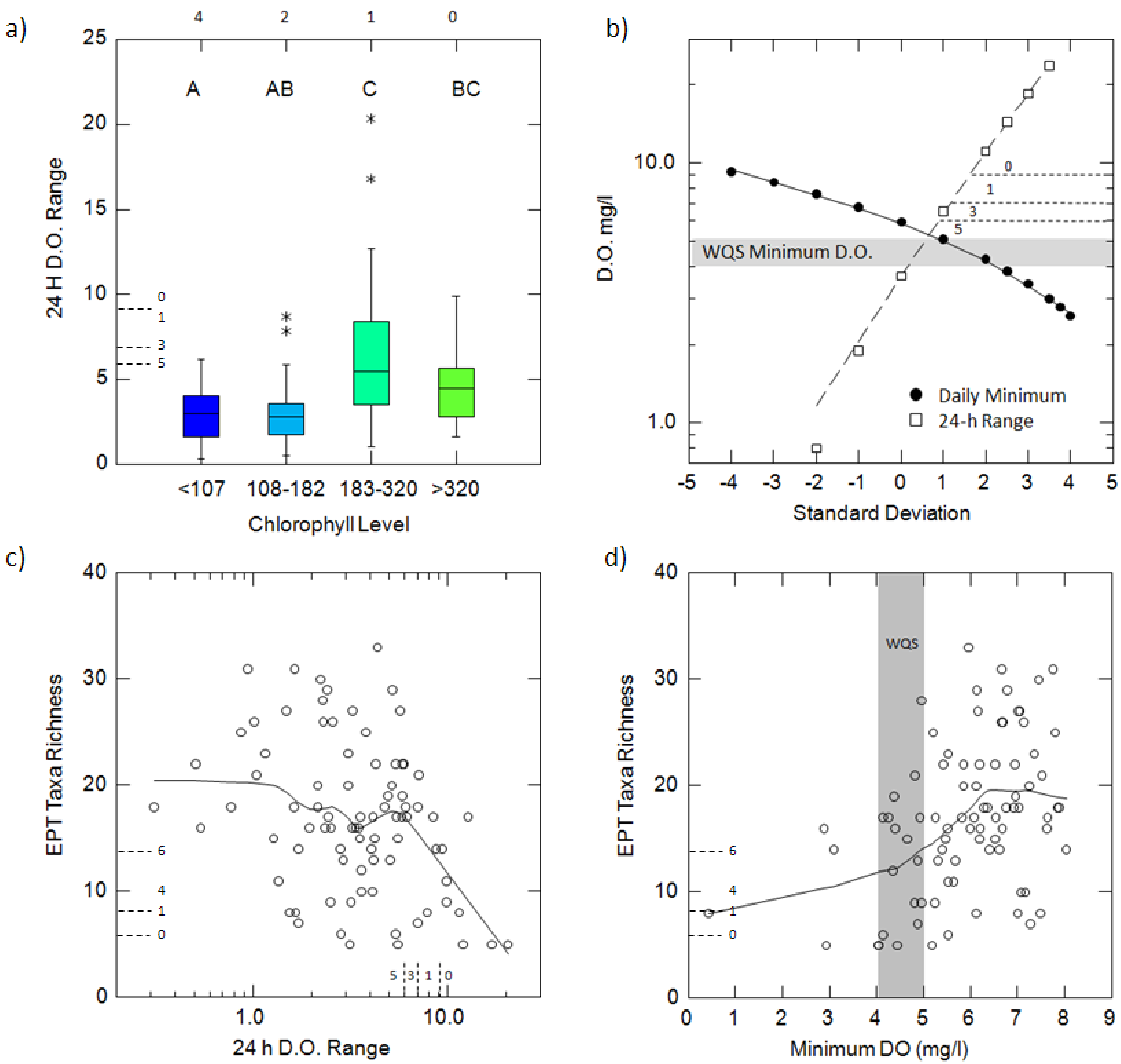

Figure 17. a) Distributions of 24 h DO range within TIC metric scoring levels of benthic chlorophyll-*a* (TIC metric scores shown on the top x-axis, concentrations on bottom x-axis). TIC metric scores for 24 h DO range are shown on the y-axis. Distributions sharing a common letter are not significantly different from each other (Kolmogorov-Smirnov test, $p<0.05$). b) DO regime in relation to deviations in the latent variable DO Stress from the SEM model. c) EPT taxa richness in relation to 24 h DO range. TIC metric scores for 24 h DO range are shown on the x-axis. Biological condition TIC metric scores are shown on the y-axis corresponding to EPT richness counts typical of excellent (i.e., metric score of 6), good (4), marginal (1) and failing (0) conditions. d) EPT richness counts plotted against minimum DO concentrations. The established WQS for minimum DO is given by the shaded area. Minimum DO concentrations falling below the WQS result in the DO regime TIC metric score of 0. Biological condition TIC metric scores are shown on the y-axis.

4.2 Testing with Recent and Historic Data: Trial With Recent Survey Data

The TIC was applied to data collected from the 2010 Biological and Water Quality Survey of Lower Great Miami River. The survey area included the lower Great Miami River (GMR) mainstem downstream from Dayton to the confluence with the Ohio River. Portions of the lower GMR are impaired by nutrient over-enrichment, as evidenced by sestonic chlorophyll-*a* concentrations exceeding 200 mg/l, 24 h DO swings in excess of 15 mg/l, frequent instances of marginal biological condition, and overt biological impairment. Several wadeable tributaries to the lower GMR were also surveyed: Clear Creek, Wolf Creek, and Bear Creek. Each receives effluent from a municipal treatment plant. The plants discharging to Clear Creek, Wolf Creek, and Bear Creek have annual flow rates of 2.1, 0.9, and 0.3 million gallons per day (mgd), respectively. In each case, phosphorus and nitrogen concentrations increase by an order of magnitude below the treatment plant outfalls (Figure 18), but with different consequences as summarized by the TIC scores calculated for each tributary (Table 12; Figure 18). In the case of Clear Creek, the nutrient load from the Springboro Wastewater Treatment Plant (WWTP) is passed on to the GMR with little or no assimilation. Wolf Creek is modestly enriched, as evidenced by high levels of benthic chlorophyll, upstream from the Brookville WWTP, then becomes overwhelmed by a combination of organic and nutrient enrichment downstream from the plant, before assimilating the load and fully recovering prior to entering the GMR. Nutrient loads from the New Lebanon WWTP to Bear Creek are assimilated without harm before reaching the GMR. Obviously, the management implications arising from these results differ for each stream.

Although the TIC scores for Clear Creek demonstrate that nutrients from Springboro are not causing impairment locally, because the load is transferred to the GMR, where nutrient impairment clearly exists, load reductions at the Springboro WWTP would likely be recommended to restore the downstream use. In the case of Wolf Creek, because the load from the Brookville plant is assimilated prior to reaching the GMR, management is directed to restore the localized impairment. Here, because the impairment starts upstream from the plant, and is caused by both organic and nutrient enrichment, the first step would be to address both upstream sources and the organic component from the plant, as that may affect restoration while reducing the nutrient load. Nutrient removal would be an option in a later cycle if needed. For the New Lebanon plant, the load is assimilated without harm, and does not currently threaten the downstream use of the GMR; therefore, under existing conditions, no action is needed.

4.3 Testing With Historic Data

Data from continuous DO monitoring were matched to biological and water data at 279 sites spanning the period from 1986 to 2006. No chlorophyll data were available, so default scores of 1 or 0 were assigned to the chlorophyll metric based on inspection of nutrient levels and the DO regime. Otherwise, metric scores were calculated as indicated in Tables 7, 9, and 10. The objectives of this exercise were twofold: first, to examine the frequency with which the TIC would label a site as either Enriched/Threatened or Impaired when the biological indicators alone indicated no impairment; and second, to examine the frequency of occurrence from historic assessments where nutrients were listed as a cause of impairment, but evidence, as given by the TIC, did not support nutrients as a cause.

Of the 279 sites, 109 sites were rated as unimpaired based on fish and macroinvertebrate assessments. Of those 109, 21.1% were rated as Enriched/Threatened by the TIC, and 4.6% were rated as impaired. Of the remaining 170 biologically impaired sites, a search of the historic assessment data base found information for 165 of those sites, of which 2.4% (4 sites) were spuriously listed as having nutrient impairment as the cause based on reassessment with the TIC. Given that pollution levels in the past were much higher, and causes, therefore, more apparent, the 2.4% error rate for spurious nutrient listings should be considered a conservative estimate (i.e., as the more overt impairments have been abated, ascribing causes to the more intransigent, multivariate, non-point related causes is a less obvious exercise). The figures based on full biological attainment, however, should extrapolate reasonably well to present conditions.

Table 12. Trophic Index Criterion scores calculated for three tributaries to the lower Great Miami River. Data are from 2010.

RM	TP	DIN	Nutrient Metric	Chl-a	Chl-a Metric	Min. DO	24 h DO Range	DO Metric	IBI	ICI	Biological Condition Metric	TIC
Clear Creek												
11.1	0.06	0.25	2	104	4	.	.	1[†]	52	44	4	11
7.6	0.03	0.59	2	126	2	3.97	9.24	0	48	42	4	8
6.9	0.72	3.54	0	155	2	6.55	4.31	5	46	36	4	11
2.5	0.55	2.28	0	83.7	4	.	.	1[†]	53	46	6	11
0.8	0.44	2.05	0	.	1[†]	.	.	1[†]	48	44	4	6
Bear Creek												
12.1	0.04	0.34	4	142	2	.	.	1[†]	46	44	4	11
9.8	0.58	3.64	0	125	2	5.75	2.91	5	44	40	4	11
7.1	0.2	0.35	1	174	2	5.14	4.38	5	42	44	4	12
0.2	0.01	0.98	2	.	1[†]	5.87	7.29	1	43	50	4	8
Wolf Creek												
16.6	0.05	0.23	2	.	1[†]	.	.	1[†]	36[ns]	24	0	4
15.3	0.06	0.68	2	296	1	4.72	3.63	5	43	22[*]	0	8
14.9	1.02	3.21	0	.	1[†]	.	.	1[†]	38[ns]	24[*]	0	2
10.4	0.31	1.11	0	210	1	6.06	3.27	5	32[*]	28[*]	0	6
2.5	0.02	0.56	2	230	1	5.73	8.43	1	51	52	6	10
0.1	0.02	0.32	4	.	1[†]	.	.	1[†]	36[ns]	46	1	7

[†]Default scores in the absence of data

[*]Biology impaired

[ns]Marginal biological condition

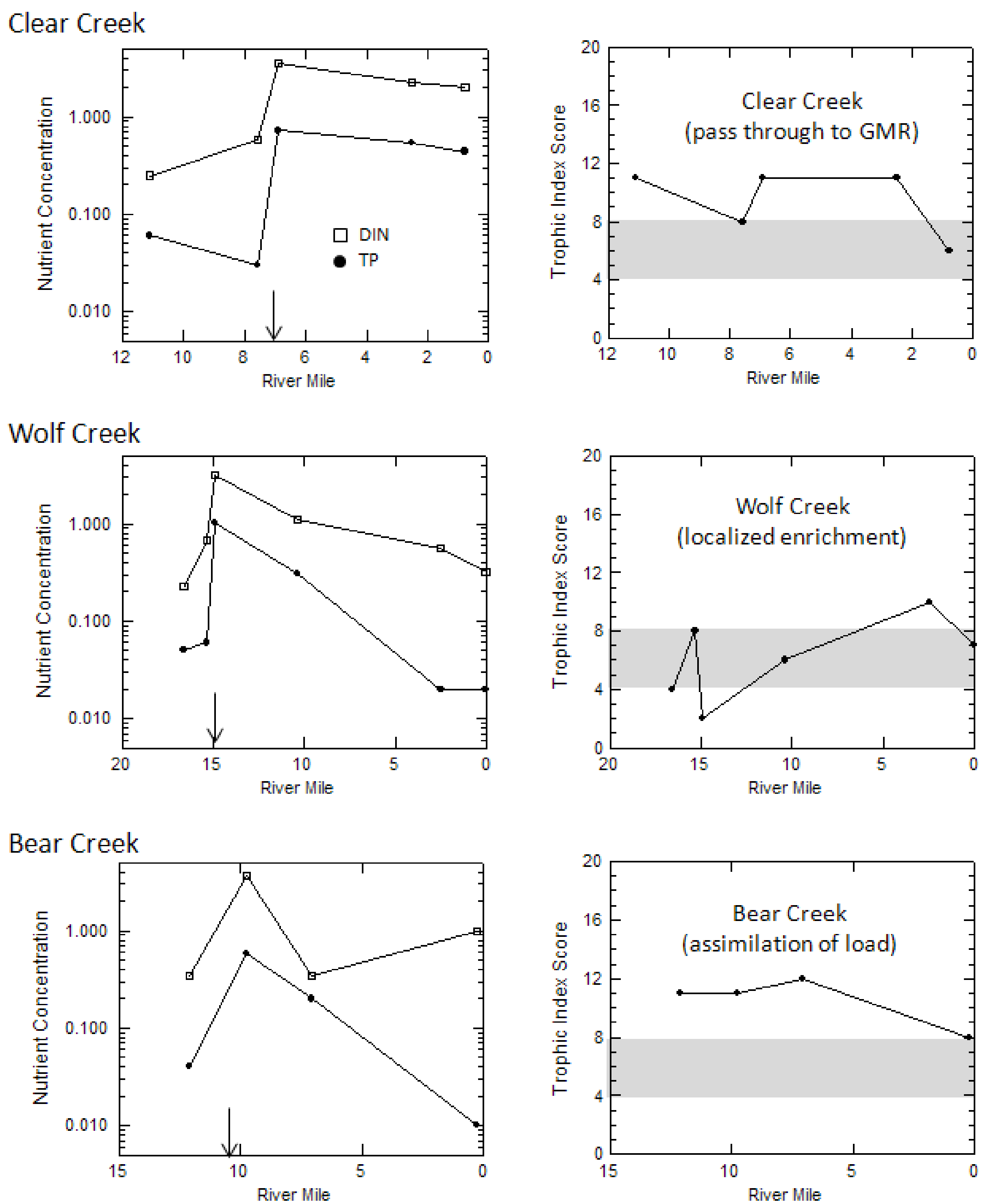

Figure 18. Nutrient concentrations and TIC scores calculated for three tributaries to the lower Great Miami River sampled in 2010. The gray-shaded region in the TIC score plots show the range of scores that indicate existing conditions threaten the use of the waterbody.

Further examination of the potential implication of the TIC on assessment outcomes was conducted using a broader slice of Ohio EPA's historic biological and water quality monitoring data. These data only have information on 2 of the 4 TIC metrics, biological condition (B) and the nutrient metric (N). TIC scores were assigned to nutrient and biological measures in the historic monitoring database, following Tables 7 and 9 (Tables 44-5 and 44-6 for TIC scoring; Working Draft Nutrient Criteria). Nutrient scores were assigned based on DIN and TP, and Biological TIC scores were assigned based on Ohio's biological criteria for each ecoregion. To reflect current conditions, rather than the more impaired conditions of the past, only data collected in 2001 and later were used.

The way the TIC is structured, at least 3 of the metrics must score in the range of 0–1 for the overall TIC to indicate nutrient impairment (4 or less). If both nutrients and biological condition score 0 or 1, then nutrient concentrations are high and IBI or ICI are impaired. Under high nutrient conditions, it is also highly probable that chlorophyll-*a* is high and the DO range fluctuates widely (Miltner 2010), and therefore the likelihood is high that the site is nutrient impaired (red cells in Table 13). Conversely, if both biological condition and nutrients have a high TIC metric value (meaning IBI/ICI are good and nutrient concentrations are low), then the TIC will exceed 8 even if benthic chlorophyll and DO fluctuations exist, and the conclusion is that the stream is acceptable without nutrient enrichment (blue cells in Table 13). With increasing biological degradation and increasing nutrient concentrations, "threatened" and "impaired" due to nutrients becomes increasingly likely (green, yellow, orange cells in Table 13). Biota that are impaired in sites that have low nutrient concentrations (gray cells in Table 13) are likely to be impaired for reasons other than nutrient enrichment.

On a statewide basis, since 2001:

- 22% of streams are biologically impaired and are highly likely to be nutrient impaired (among other causes).
- 20% of streams are biologically impaired; they may be nutrient threatened but are unlikely to be nutrient impaired; biological impairment may be due to other causes.
- 30% are biologically unimpaired but may be nutrient threatened or nutrient impaired. Note that this figure comports well with the 25.7% estimated from data where information from continuous oxygen monitoring were available.
- 28% are biologically unimpaired and highly unlikely to be nutrient impaired or threatened.

Table 13. Distribution of Ohio stream samples, 2001 – 2010, among TIC metric categories (biological and nutrient), by Ecoregion. Colors indicate categories of the likelihood of nutrient impairment given that 2 metrics (DO and benthic chl-a) are unknown for the historic, statewide data set. Color indications shown in Legend.

		Nutrient TIC score				
ECOREGION	**Bio TIC score**	**0**	**1**	**2**	**4**	**Row Totals**
Eastern Cornbelt Plains (55)	0	6%	13%	8%	1%	28%
	1	3%	7%	2%	0%	13%
	4	4%	19%	8%	2%	35%
	6	3%	14%	6%	1%	24%
Total		16%	54%	25%	4%	604
Huron-Erie Lake Plain (57)	0	1%	19%	2%	0%	22%
	1	5%	9%	1%	0%	15%
	4	26%	23%	9%	2%	60%
	6	1%	0%	0%	1%	2%
Total		33%	51%	12%	4%	81
Erie-Ontario Lake Plain (61)	0	7%	12%	10%	3%	31%
	1	3%	6%	4%	3%	15%
	4	5%	13%	14%	6%	38%
	6	2%	6%	6%	2%	16%
Total		16%	37%	33%	14%	400
Western Allegheny Plateau (70)	0	0%	3%	8%	17%	29%
	1	0%	3%	2%	6%	12%
	4	0%	7%	7%	14%	29%
	6	0%	6%	7%	17%	31%
Total		2%	19%	24%	55%	442
Interior Plateau (71)	0	8%	6%	1%	0%	15%
	1	0%	4%	0%	0%	4%
	4	7%	33%	6%	3%	49%
	6	6%	18%	6%	3%	32%
Total		21%	61%	13%	6%	72
Statewide	0	4%	10%	8%	6%	28%
	1	2%	6%	3%	3%	13%
	4	5%	15%	9%	7%	36%
	6	2%	9%	6%	6%	23%
Total		13%	40%	26%	21%	1599

Table 13 Legend.

Legend
Unimpaired, not nutrient "threatened"
Biologically unimpaired, low probability of nutrient "threatened" status
High probability of "threatened" status, low to moderate probability of nutrient impairment
Biologically impaired, moderate probability of nutrient impairment
Biologically impaired, high probability of nutrient impairment
Biologically impaired but not nutrient impaired (other causes)

5. Programmatic Implementation

5.1 Background on Ohio EPA's Biological Survey Program

Biological information is the basis for determining the status of beneficial aquatic life uses in Ohio. Although Ohio's original WQS included a single general aquatic life use classification, in 1978 Ohio EPA adopted a tiered classification scheme for different aquatic life uses. In this scheme, waters are classified based on the chemical, physical, and biological variability inherent in natural aquatic ecosystems. Ohio has established numeric standards for biological quality, and these standards are tiered according to aquatic life uses. Ohio EPA has been intensively monitoring the condition of Ohio's surface waters since the late 1970s, with 20,065 samples taken at 9,250 sites between 1978 and 2005 (Figure 19). Each year, Ohio EPA conducts biosurveys in 5–6 study areas, with a total of 250–300 sampling sites (Yoder and Rankin 1999). Biosurveys gather information on chemical, physical, sediment, and habitat quality, as well as biological condition. Biosurvey information is used to determine the extent to which biological criteria are met; identify stressors in cases where the criteria are not met; determine whether the habitat classification criteria assigned to each waterbody are appropriate and attainable; and determine whether any changes in biological, chemical, or physical indicators have occurred since earlier measurements, particularly before and after the implementation of point source pollution controls or best management practices for nonpoint sources (Yoder and Rankin 1999).

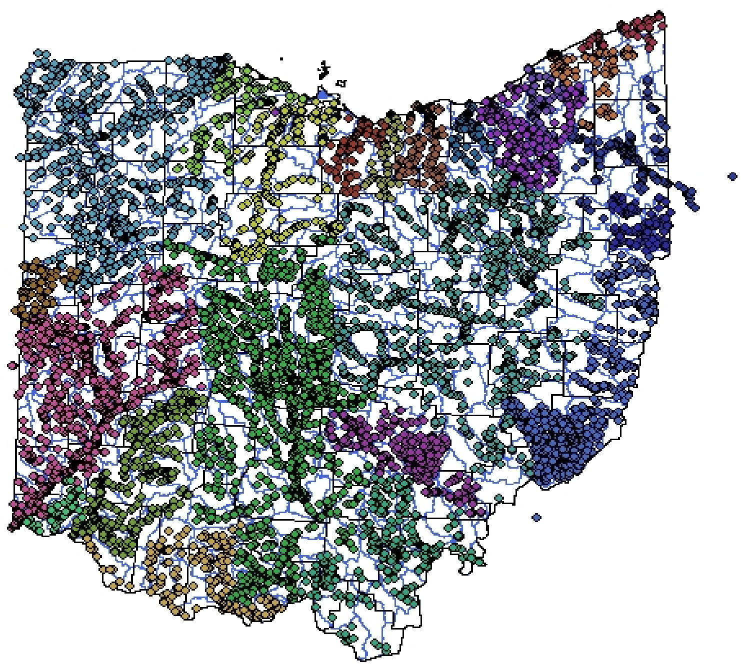

Figure 19. Ohio EPA's Biological Sampling Locations: 1978 - 2005.

In Ohio's WQS, aquatic life use designations are assigned to individual waterbody segments based on the potential to support the use according to the narrative and numerical criteria in the absence of existing stressors, rather than observation of attainment of criteria alone (Yoder and Rankin 1995). This provides the basis for improving and rehabilitating degraded aquatic systems. Ohio EPA uses a standardized classification hierarchy to set measureable goals for specific surface waterbodies. However, while the 1978 tiered uses were based on ecological attributes, the criteria associated with them were entirely chemical and physical. In 1980, Ohio EPA developed narrative biocriteria for the tiered uses. The narrative criteria adopted in 1980 consisted of narrative expressions and numeric biological index guidelines which reflected more directly the measurable components of the aquatic life use designations. The narrative classification system consisted of assigning performance categories such as exceptional (meets the EWH use), good (meets the WWH use), fair, poor, and very poor (the latter three fail to attain the minimum CWA goal for aquatic life use). The purposes of the narrative classification system were to provide a systematic basis for assigning aquatic life uses to surface waters, and to provide a standardized approach to determining the magnitude and severity of impairments to the aquatic biota. Numeric indices used to help define the narrative classification system were comprised of single-dimension measures such as taxa richness, Shannon diversity index, and the Index of Well-Being (Iwb). Attainable expectations for a suite of narrative community attributes were based on Ohio EPA experience with sampling approximately 200 sites statewide (Yoder and Rankin 1995).

In 1990 Ohio EPA adopted numeric biocriteria, thus formalizing bioassessment in Ohio's programs. These criteria are based on measurable characteristics of fish and macroinvertebrate assemblages (e.g., species richness, key taxonomic groupings, functional guilds, environmental tolerances, and organism condition), wherein numeric expectations are defined by data collected from more than 350 reference sites (OEPA 1987a, b; 1989a, b; Yoder 1989; Yoder and Rankin 1995). Ohio EPA uses three multi-metric indices to assess fish and macroinvertebrate communities: the IBI, the Modified Index of Well-Being (MIwb), and the ICI. A biotic index is a numeric representation of a measured assemblage, wherein the numeric index is calibrated against a set of reference sites, and validated against known stressor gradients, such that the numeric score for a given site is a representation of where that site is positioned along a biological condition gradient ranging from fully natural to entirely altered and degraded.

Numeric standards for aquatic life represent the degree of biological integrity that can reasonably be expected given contemporary land uses, in a framework that includes a provision to change the biocriteria in response to any future improvements in conditions at reference sites. This provides a realistic framework against which to evaluate contemporary environmental management and restoration efforts (Yoder and Rankin 1999). For instance, the numeric biocriteria for the warmwater habitat designation is based on the 25th percentile value of reference site scores by index (IBI, MIwB, ICI), site type (large, medium, or headwater stream), and ecoregion—this provides numeric biocriteria that vary by ecoregion in accordance with the narrative definition and the reference site results for each site type (Yoder and Rankin 1995).

Ohio classifies waters into seven aquatic life designated uses, with associated chemical, physical, and biological criteria (OAC 3745-1-07):

- Coldwater Habitat
- Exceptional Warmwater Habitat
- Seasonal Salmonid Habitat
- Warmwater Habitat
- Limited Warmwater Habitat
- Modified Warmwater Habitat
- Limited Resource Waters

5.2 Integration with Other Programs

Biological information, in combination with chemical and physical data, provides a direct measure of ecological integrity in a waterbody. In addition to setting WQS standards, Ohio's bioassessment information supplies critical information to all water quality management programs (Ohio EPA, 1987a, Yoder and Rankin 1995, Yoder and Rankin 1996, Yoder and Rankin 1999). Applications include:

- NPDES permitting—Ohio uses biological information to ensure that effluent limits in NPDES permits result in meeting water quality goals. Biological information can also be used in enforcement decisions and to support litigation. A discussion of integrating biological monitoring for nutrient enrichment into the NPDES program can be found in this chapter.
- Review and modification of designated uses for waterbodies—Ohio EPA has upgraded many stream miles from lower to higher categories (e.g., WWH to EWH; Limited to WWH) based on actual attainment results following improvements in discharges or restoration.
- Ohio Water Resource Inventory—Information on aquatic life use attainment is used to develop Ohio's biennial CWA section 305(b) report. Standardized biological and chemical monitoring has resulted in a database housing a long period of record that can determine trends in water quality, and provide empirical evidence for stressor-response relationships.
- Nonpoint source assessment and management (CWA section 319)—Biological criteria provides an environmental endpoint that is combined with other environmental information (e.g., data on land use, riparian zones, sub-ecoregional characteristics) to assess and manage nonpoint source impacts on water quality.
- Evaluation of wet weather flow impacts—Bioassessments can be used to measure a biological community's response to impacts on the physical habitat such as sedimentation from stormwater runoff and physical habitat alterations from dredging, filling, and channelization. Biological data has been used to assess how changes in urban landscapes can impact changes in stormwater and combined sewer overflows, and those data can provide useful information to inform management and remediation efforts.
- CWA section 401 certifications— Under CWA section 401, no federal agency can issue a permit or license that may result in a discharge to CWA jurisdictional waters without certification from the state water quality agency that the discharge would be consistent with the state's WQS and other water quality goals. Biological information provides support for assessing habitat manipulation and degradation and in granting or denying CWA section 401 certifications.
- Other, non-CWA uses—Biological survey data have been used to provide information on rare, threatened, and endangered species. These data are used to show trends in degradation and recovery of Ohio's fauna. Such data have also been used in the management and assessment of fisheries.

5.3 Programmatic Implementation of the TIC and Numeric Nutrient Criteria

Unlike toxic substances, nutrients do not simply kill aquatic organisms by direct action, so nutrient criteria cannot be built on the assumptions that less is always better and that a simple threshold will protect all waters. More importantly, this document, as well as EPA reports and the scientific literature, have demonstrated that manifestation of harmful consequences of nutrient enrichment depend on site-specific and local conditions. Ohio EPA recognizes that site-specific nutrient criteria for every single waterbody in the state is impractical in the extreme, and also seeks proactive, protective nutrient criteria that would avoid only taking action after damage has been done. The TIC, as a proposed criterion, uses multiple lines of evidence to assess the ordinal risk of nutrient enrichment causing harm to the valued endpoints of biological integrity and DO. The multiple lines of evidence take into account site-specific information captured in Ohio's monitoring program, as demonstrated by the analyses and model results in this document. The TIC provides for implementation as protective criteria, yet it is not a simple threshold of DIN and TP.

Because Ohio EPA has a robust monitoring program, supplying data for the TIC is almost a matter of routine. The only added monitoring component is collection of benthic chlorophyll. When a waterbody is positioned on the nutrient continuum, then comparison to the numeric nutrient criteria can be interpreted in context, and applied if necessary. Note that the numeric criteria for TP and DIN differ from their respective TIC scoring thresholds, as the TIC thresholds are simply a four-part division of the log-scale range, and reflect how close concentrations are to the criterion, and hence the probability of failing the WWH criterion. The numeric criteria represent the synthesis of all available information from the nutrient study, and especially from the results of logistic regression. These criteria offer protection for existing high quality waters and provide realistically achievable targets where impairment is documented. To illustrate this point, a comparison of biological response against ammonia nitrogen and total phosphorus in light of existing and proposed criteria is helpful.

Figure 20 plots IBI and ICI scores against ammonia nitrogen and phosphorus for small streams in the Eastern Cornbelt Plains Ecoregion (ECBP), and overlays existing criteria[5] for ammonia nitrogen and proposed criteria for total phosphorus. The position of the chronic ammonia criterion relative to biological response along the ammonia gradient, compared to the position of the restoration criteria relative to biological response against the total phosphorus gradient, suggests that phosphorus criteria are at least as protective, if not more so, than the existing ammonia criterion in a relative sense. Although the chronic standard for ammonia nitrogen may not appear sufficiently protective, that standard has been used to great effect over the last 25 years—advanced wastewater treatment and permit limits based on that standard are largely the reason impairment in Ohio's medium and large rivers has been reduced from 79 to 11 percent. The 0.3 mg/l TP criterion for NPDES permit limits and wasteload allocations is analogous to the ammonia standard in that it is a workable starting point for waters presently over-enriched by point sources. Given the strong connection between habitat quality, land use, and the expression of excess nutrients as over-enrichment, the 0.130 mg/l TP criterion is necessary where habitat quality is marginal or degraded. The 0.130 mg/l TP criterion also offers a secondary target, based on iterative permit cycles, should a 0.3 mg/l initial target prove ineffectual at achieving restoration. This iterative approach recognizes the greater level of uncertainty in the biological response to nutrients compared to conventional pollutants. In these latter two regards, implementation of nutrient criteria fall outside the traditional regulatory and programmatic framework.

[5] Ammonia criteria are based on a combination of temperature and pH. For headwater and wadeable streams in the ECBP, that translates to a chronic criterion of ~ 1.0 mg/l more often than not.

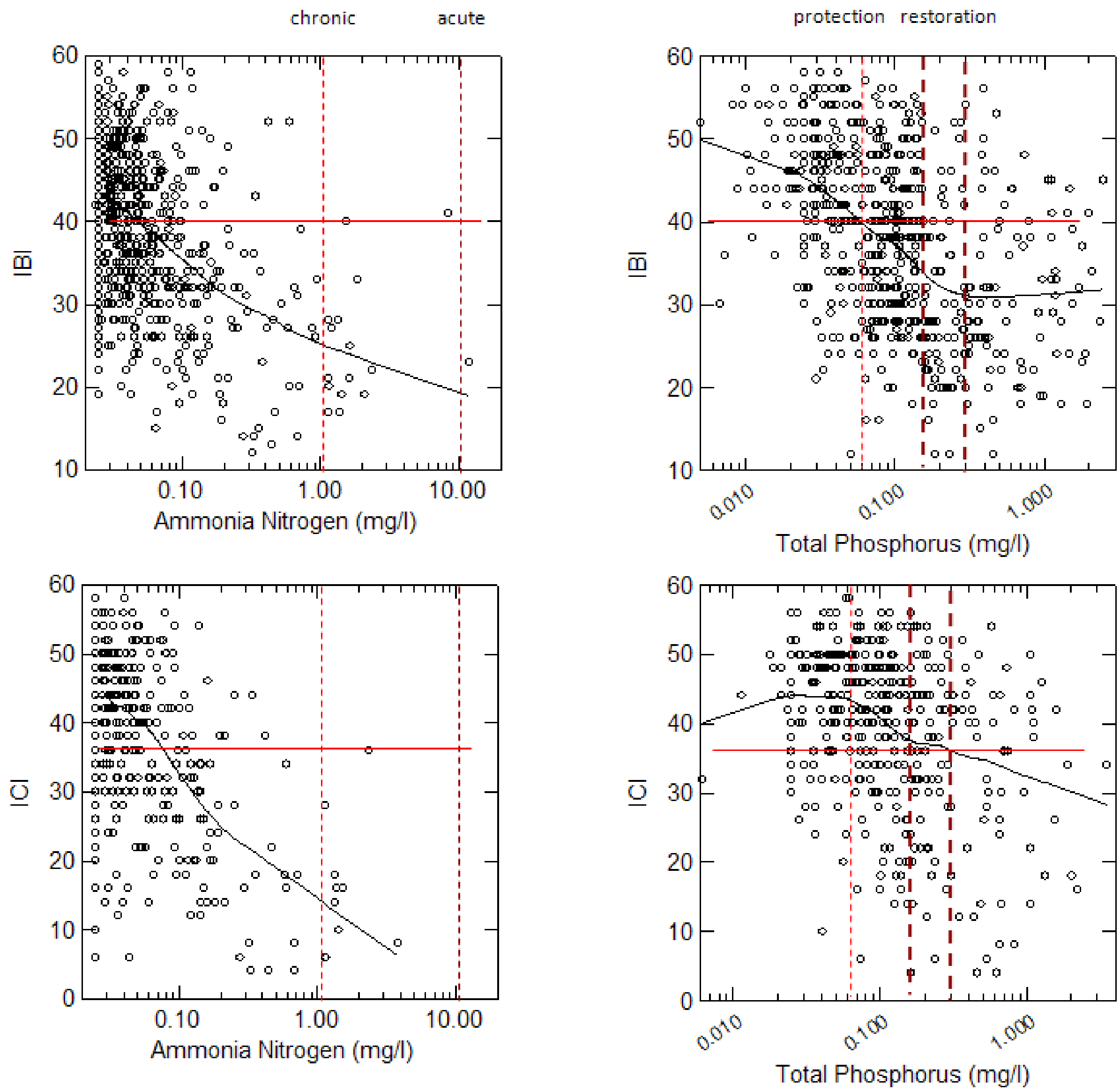

Figure 20. Response of biological assemblages in relation to ammonia nitrogen and total phosphorus concentrations and in light of existing and proposed numeric criteria (vertical dashed lines). Data are for small streams in the ECBP, and span the years 1982–2010. The solid, red horizontal line joining the y-axis in each plot shows the respective biological criterion. The solid, black line following the local central tendency in each plot is from LOWESS (q=0.5).

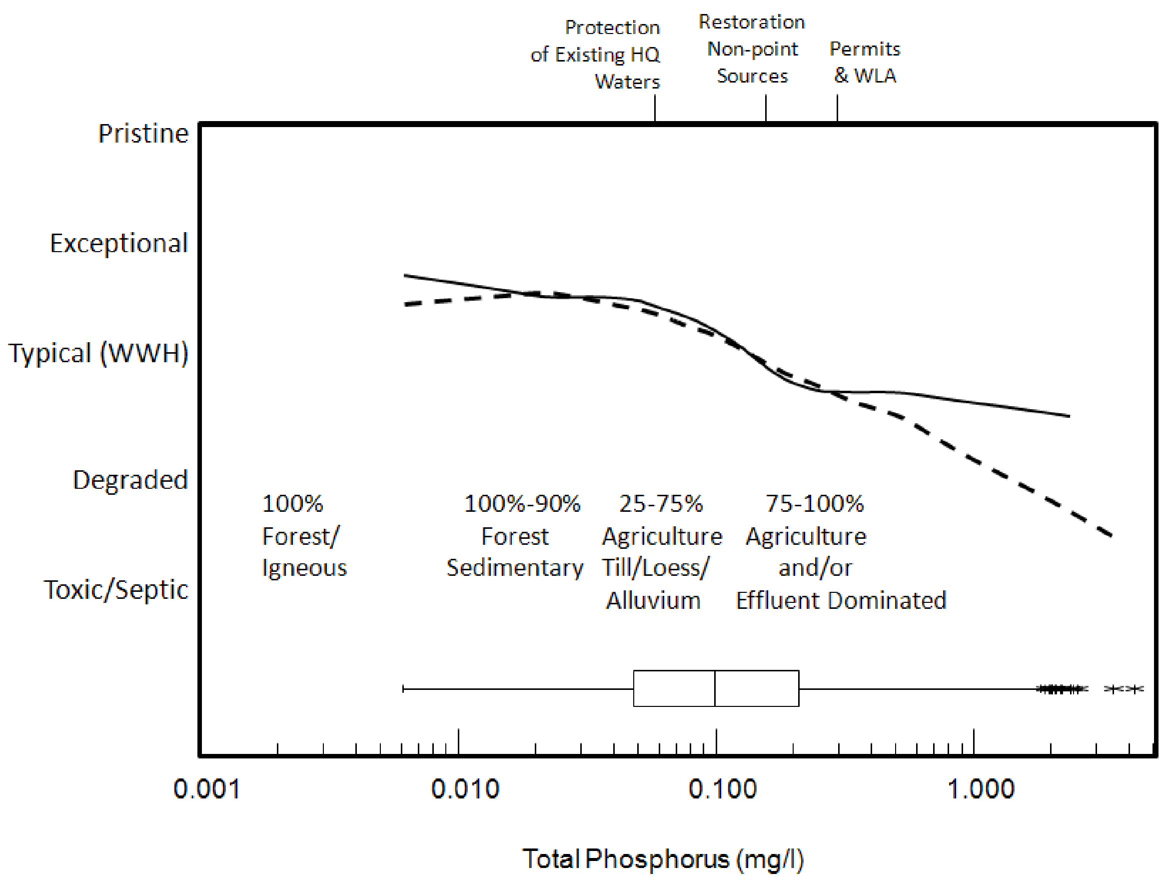

Figure 21. Nutrient criteria for TP in the context of the bio-condition gradient (y-axis) and land use. The box plot shows the distribution of TP concentrations in the ECBP ecoregion. Land use categories are positioned over the range of phosphorus concentrations typical of the category (i.e., 100% forest cover over igneous bedrock is shown for perspective, though that clearly is not found in the ECBP). The solid and dashed lines are the LOWESS trend lines from Figure 19, and shown to scale (IBI solid, ICI dashed).

From the standpoint of reasonable potential, projected ammonia concentrations exceeding 1.0 mg/l clearly have the potential to do harm. Phosphorus concentrations exceeding 0.3 mg/l are more than likely to result in non-attainment, but unlike ammonia, there is reasonable uncertainty, as evidenced by the scatter plots (Figure 20). Hence the need for additional evidence and context before invoking reasonable potential. Again, most of that evidence is collected as a matter of routine during biological and water quality surveys. Broader context can be had by examining the local habitat quality, longitudinal profiles of water quality and biological measures down the run of the river, proximity of other stressors, and land use in the surrounding catchment (e.g., see Figure 21). Lastly, coefficients in the structural equation model (Figure 8) can be used to project future conditions for the purposes of estimating TIC scores.

Regarding nitrogen, an across-the-board 3.0 mg/l criterion for dissolved inorganic nitrogen is proposed, in recognition that nutrient co-limitation exists and was evident in the nutrient data. However, with respect to point sources, phosphorus removal will invariably be the cost effective alternative, as evidenced by the discussion on pages 26-27 and demonstrated in Figures 11 and 12. Therefore, DIN limits for point sources are not likely to be a matter of routine in cases where impairment is demonstrated, but may be imposed if TP control alone does not affect restoration.

5.4 TMDL/WLA and Calculation of WQBELs

[work in progress – the following is boilerplate from the draft rule as a place holder]

(A) For discharges of nutrients (total phosphorus and dissolved inorganic nitrogen) to flowing receiving waters, the wasteload allocation (WLA) shall be calculated using the following mass balance equation:

$$\frac{WQC(Q_{eff} + Q_{up}) - Q_{up}(WQ_{up})}{Q_{eff}}$$

Where:

WQC = water quality criterion as established in table 44-2 of rule 3745-1-44 of the Administrative Code;

Q_{eff} = effluent design flow as established in rule 3745-2-05 of the Administrative Code;

Q_{up} = per cent of the upstream design flow as established in paragraphs (C) and (D) of this rule; and

WQ_{up} = background water quality as established in rule 3745-2-05 of the Administrative Code.

(B) Where sufficient data exists, alternative modeling methods (such as QUAL2K) that simulate nitrogen, phosphorus, DO and chlorophyll at a minimum, will be used. Similar modeling methods will be used where possible to determine appropriate load allocations for new discharges to ensure they are appropriate and protective of water quality criteria.

(C) The following stream design flows shall be used to determine WLAs to maintain water quality criteria for nutrients:

(1) May to November: summer stream flow exceeded eighty per cent of the time; and

(2) December to February: winter stream flow exceeded eighty per cent of the time.

(D) The WLAs shall use the per cent of stream design flow contained in paragraphs (A)(2)(a) to (A)(2)(c) of rule 3745-2-05 of the Administrative Code. The director may determine design flows for streams that are impacted by reservoirs or other physical alternations by taking into account relevant site-specific factors. Stream design flows for such impacted stream segments shall be established at levels that ensure protection of designated uses. Alternative flows or seasons may be used if the director determines that the flow or season is as protective as those listed in paragraph (C) of this rule.

(E) Multiple discharges. When the director determines that it is necessary to consider multiple discharges in a WLA, the loading capacity may be distributed among discharges using a method deemed appropriate by the director based on site-specific considerations.

5.5 NPDES Permits–Reasonable Potential to Contribute to WQS excursions

Under federal NPDES program rules, permits must control all pollutants that have the reasonable potential to cause or contribute to excursions of water quality criteria [40 CFR 122.44(d)]. Permitting authorities, including delegated states, must have procedures for determining reasonable potential that account for existing controls on point and non-point sources of pollutants, the variability of pollutants in a regulated discharge, and stream dilution where appropriate. When a permitting authority determines that a discharge has reasonable potential for a given pollutant, the permit must contain a limit for that pollutant. In this situation, when a discharger is found to have the reasonable potential to cause or contribute to an excursion of the TIC, the permit would include limits on the nutrient(s) causing the excursion–phosphorus and/or DIN.

The TIC accounts for controls on point and nonpoint sources in several ways. At the most fundamental level, nonpoint sources are counted in the background (upstream) concentrations used in the wasteload allocation. For a critical flow allocation, these would be median or mean upstream concentrations as defined in OAC Rule 3745-2-05. The background concentration could be adjusted if nonpoint source controls specified in a TMDL would project a lower background concentration.
Point source controls can also be incorporated into background concentrations. If the point sources are located relatively close to each other, or impacts extend from one discharger to another, the point sources can be allocated together, with each given its share of the available stream load, similar to TMDL calculations. Allocations may be done on a stream-segment or watershed basis.

Effluent variability is considered in the Projected Effluent Quality (PEQ) measures used to assess point source discharges. PEQ is a statistical calculation, and considers mean effluent values and variability. Ohio EPA determines effluent PEQ values through the Implementation of Water Quality Standards Rules, OAC 3745-2; we would use these same procedures for phosphorus and DIN in NPDES effluents.

5.6 Uses of the TIC in Determining Reasonable Potential

Many NPDES permittees discharge phosphorus and DIN. These discharges need to be evaluated to determine whether they have the reasonable potential to contribute to excursions of WQS. As the TIC is the criterion, TIC values measured downstream of a discharger and projected to the 80th percentile flow are used to determine reasonable potential. If reasonable potential exists, then Ohio EPA would set limits for phosphorus and/or DIN in response.

Ohio EPA is proposing to use an 80th percentile exceedance flow in stream modeling for the TIC, and TP and DIN. The 80th percentile flow was selected to be representative of common, low-flow conditions. Enrichment impacts on a waterbody are chronic impacts, reflecting seasonal or long-term loading trends. As a result, a flow such as the 80th percentile more accurately reflects these conditions than very rare occurrences such as the 7Q10 or 1Q10 flows used for toxic pollutants. For existing discharges, use of the TIC in reasonable potential determinations would be similar to the methods for determining compliance with WQS mentioned above. Metrics would be evaluated using available data or default values for affected downstream segments. If the modeled TIC score is in the impaired or threatened category, reasonable potential would exist, and limits based on the WLA would be included in the NPDES permit.

TIC scores downstream from a new or expanding discharger would be projected using modeling techniques. For expanding dischargers, this may involve using existing TIC data projected to new conditions; for new discharges models will need to incorporate factors from the TIC to project nutrient effects at the 80^{th} percentile exceedance flow.

Most permits that establish nutrient limits under this framework will have compliance schedules that allow time to meet the new limits. These schedules will contain a requirement to meet the wasteload allocation, and may include an option for water quality trading, or adaptive management, to meet the numeric nutrient standard(s). We anticipate that many dischargers will choose the adaptive management alternative if available.

The trading/adaptive management option would require meeting interim effluent limits not greater than 1.0 mg/l total phosphorus and 10 mg/l DIN as monthly averages. It would require participation in a water quality trading program, with actions to be implemented and annual reporting to Ohio EPA as conditions of the permit. Measurement of certain TIC metrics at downstream locations would also be included in the permit.

Ohio EPA would be able to terminate the trading program and require compliance with WLA limits if the permittee failed to comply with trading-related requirements included in its NPDES permit, if there was a failure by participants to implement actions in the approved trading plan, or if the trading program failed to generate adequate credits to meet water quality based effluent limits. The trading option is included because effluent loadings of phosphorus and DIN are not the only factors involved in nutrient impact. As mentioned earlier, non-point source loadings and stream habitat characteristics are also significant factors. The trading option is a way to attain the numeric nutrient standard(s) by the most cost-effective means.

5.7 Watershed Assessments, NPS Recommendations

Aquatic life use and water quality assessments of Ohio's surface waters are conducted annually. Assessments are based on systematic surveys of whole catchments that typically range in size from 200–500 mi^2, or longitudinal surveys of large river mainstem segments. Sampling location density within a catchment is at approximately one site per 5 mi^2 of drainage area, supplemented by targeted sampling in proximity to NPDES discharge locations, and known or potential sources of localized pollution. Routine sampling includes water chemistry, biological quality, habitat quality, and sediment chemistry. Additionally, several to a dozen sites are selected systematically within the stream network according to fixed increments of drainage area for monthly monitoring of water chemistry to capture seasonal trends and loadings. Samples for benthic and sestonic chlorophyll are collected from a subset of sites, dictated by proximity to point sources, or known or suspected nonpoint sources. Automated DO sensors are deployed at chlorophyll sampling locations. Data from the watershed surveys are supplemented with information on loadings from NPDES monthly operating reports, as well as categorical characterizations of land use, and non-point sources (e.g., densities of CAFOs, home sewage treatment systems, percent impervious cover, etc.). Analysis of the resulting data and information are synthesized into a technical support document and incorporated into a TMDL study. The end result is a condition assessment of the watershed that includes a characterization of causes and sources of impairment, as well as recommended load reductions and pollution abatements needed to restore beneficial uses.

Within the framework of the watershed assessment, the TIC will inform both condition status and causal determinations, the former by positioning waterbody segments on the enrichment gradient, and the latter by providing evidence for nutrients as a proximate cause of non-attainment. For waterbodies identified as nutrient impaired by non-point sources, this will allow focused management actions aimed at abating the localized impairment, rather than applying broadly prescriptive measures. On a watershed scale, the TIC, in conjunction with loadings estimates, will help determine the amount and kinds of broadly prescriptive management actions necessary for over-all load reduction.

5.8 Protection of Downstream Waters

Large rivers tend to carry higher concentrations of soluble minerals and sestonic algae (Van Nieuwenhuyse and Jones 1994) compared to smaller streams as a consequence of erosion, deposition, retention, and sunlight, and consequently are relatively more enriched. The upshot of this is that nutrient thresholds and numeric criteria derived from small streams and headwaters are inherently protective of the larger downstream reaches. Furthermore, given that loadings from diffuse sources, especially agriculture, account for the majority of transported nutrients (USGS 2010), load allocations based on the nutrient targets developed from small streams are, axiomatically, necessary for protecting downstream waters. That said, robust monitoring is necessary to not only understand the fate and transport of locally derived nutrients, but the condition of downstream receiving waters, as evidenced by the example for the lower GMR outlined previously, and clearly extends to lakes. Numeric nutrient criteria and assessment methods for lakes have been proposed by Ohio EPA that will dictate the quality of influent waters where necessary based on TMDLs.

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Appendix A. Results from logistic regression.

```
Logistic Regression Results for Fish IBI 0<WWH, 1>WWH


  REF              1164
  RESP              884
Total      :       2048

PHOSPHORUS MODEL
Log Likelihood:  -1159.90104
    Parameter                Estimate         S.E.      t-ratio      p-value
  1 CONSTANT                 -1.67746      0.32055     -5.23311      0.00000
  2 QHEI                      0.06716      0.00412     16.31860      0.00000
  3 TP                       -1.24038      0.11687    -10.61348      0.00000
                                                  95.0 % bounds
    Parameter              Odds Ratio        Upper        Lower
  2 QHEI                      1.06947      1.07813      1.06088
  3 TP                        0.28927      0.36374      0.23005
Log Likelihood of constants only model = LL(0) =  -1400.36472
2*[LL(N)-LL(0)] =    480.92736 with 2 df Chi-sq p-value = 0.00000
McFadden's Rho-Squared =      0.17172
Deciles of Risk

Records processed: 2048
Sum of weights =   2048.00000
                            Statistic      p-value           df
Hosmer-Lemeshow               5.72055      0.67850      8.00000


NITROGEN MODEL
Log Likelihood:  -1207.62230
    Parameter                Estimate         S.E.      t-ratio      p-value
  1 CONSTANT                 -2.43785      0.35350     -6.89625      0.00000
  2 QHEI                      0.06822      0.00402     16.96804      0.00000
  3 DIN                      -0.54758      0.09790     -5.59304      0.00000
                                                  95.0 % bounds
    Parameter              Odds Ratio        Upper        Lower
  2 QHEI                      1.07060      1.07907      1.06219
  3 DIN                       0.57835      0.70069      0.47737
Log Likelihood of constants only model = LL(0) =  -1402.89936
2*[LL(N)-LL(0)] =    390.55412 with 2 df Chi-sq p-value = 0.00000
McFadden's Rho-Squared =      0.13920
Deciles of Risk

Records processed: 2052
Sum of weights =   2052.00000
                            Statistic      p-value           df
Hosmer-Lemeshow               5.33214      0.72156      8.00000
```

```
Logistic Regression Results for Macroinvertebrate ICI 0<WWH, 1>WWH

REF               577
RESP              410
Total      :      987
PHOSPHORUS MODEL
Log Likelihood:   -587.81857
    Parameter                Estimate         S.E.      t-ratio      p-value
  1 CONSTANT                 -0.45632      0.46659     -0.97798      0.32808
  2 QHEI                      0.04981      0.00523      9.51959      0.00000
  3 TP                       -1.20616      0.17477     -6.90160      0.00000
                                                   95.0 % bounds
    Parameter              Odds Ratio        Upper        Lower
  2 QHEI                      1.05107      1.06190      1.04034
  3 TP                        0.29934      0.42163      0.21252
Log Likelihood of constants only model = LL(0) =   -669.93991
2*[LL(N)-LL(0)] =    164.24268 with 2 df Chi-sq p-value = 0.00000
McFadden's Rho-Squared =      0.12258
Deciles of Risk

Records processed: 987
Sum of weights =    987.00000
                            Statistic      p-value           df
Hosmer-Lemeshow               9.37588      0.31159      8.00000


NITROGEN MODEL
Log Likelihood:   -606.64250
    Parameter                Estimate         S.E.      t-ratio      p-value
  1 CONSTANT                -10.98987      2.12190     -5.17926      0.00000
  2 QHEI                      0.05010      0.00512      9.78228      0.00000
  3 DIN                       5.69232      1.47699      3.85400      0.00012
  4 DIN2                     -0.96295      0.25599     -3.76174      0.00017
                                                   95.0 % bounds
    Parameter              Odds Ratio        Upper        Lower
  2 QHEI                      1.05138      1.06199      1.04088
  3 DIN                     296.58234   5362.45642     16.40313
  4 DIN2                      0.38176      0.63051      0.23115
Log Likelihood of constants only model = LL(0) =   -670.81771
2*[LL(N)-LL(0)] =    128.35041 with 3 df Chi-sq p-value = 0.00000
McFadden's Rho-Squared =      0.09567
Deciles of Risk

Records processed: 988
Sum of weights =    988.00000
                            Statistic      p-value           df
Hosmer-Lemeshow               1.86778      0.98480      8.00000
```

```
Logistic Regression Results for EPT Taxa Richness 0≤10 taxa, 1>10 taxa

  REF              514
  RESP            1693
Total      :      2207

PHOSPHORUS MODEL
Log Likelihood:  -1142.14571
    Parameter               Estimate         S.E.      t-ratio      p-value
  1 CONSTANT                -4.34237      0.35278    -12.30887      0.00000
  2 QHEI                     0.03917      0.00403      9.70892      0.00000
  3 TP                       0.33615      0.11154      3.01378      0.00258
                                                 95.0 % bounds
    Parameter             Odds Ratio        Upper        Lower
  2 QHEI                     1.03994      1.04820      1.03175
  3 TP                       1.39954      1.74151      1.12473
Log Likelihood of constants only model = LL(0) =  -1197.85191
2*[LL(N)-LL(0)] =    111.41242 with 2 df Chi-sq p-value = 0.00000
McFadden's Rho-Squared =      0.04651
Deciles of Risk

Records processed: 2207
Sum of weights =   2207.00000
                            Statistic      p-value           df
Hosmer-Lemeshow               9.01796      0.06065      4.00000


NITROGEN MODEL
Log Likelihood:  -1126.30104
    Parameter               Estimate         S.E.      t-ratio      p-value
  1 CONSTANT               -10.78678      1.74280     -6.18934      0.00000
  2 QHEI                     0.03748      0.00402      9.32485      0.00000
  3 DIN                      4.46851      1.19748      3.73159      0.00019
  4 DIN2                    -0.67015      0.20483     -3.27176      0.00107
                                                 95.0 % bounds
    Parameter             Odds Ratio        Upper        Lower
  2 QHEI                     1.03820      1.04641      1.03005
  3 DIN                     87.22643    911.89724      8.34354
  4 DIN2                     0.51163      0.76438      0.34246
Log Likelihood of constants only model = LL(0) =  -1198.91134
2*[LL(N)-LL(0)] =    145.22061 with 3 df Chi-sq p-value = 0.00000
McFadden's Rho-Squared =      0.06056
Deciles of Risk

Records processed: 2211
Sum of weights =   2211.00000
                            Statistic      p-value           df
Hosmer-Lemeshow               5.37978      0.25050      4.00000
```