



# Nutrient Breakpoints and Identifying Nutrient Reference Sites in Indiana

Shivi Selvaratnam<sup>1</sup> and Jeff Frey<sup>2</sup>

<sup>1</sup> Indiana Department of Environmental Management, Office of Water Quality,  
Indianapolis, IN

<sup>2</sup> USGS Indiana Water Science Center, Indianapolis, IN



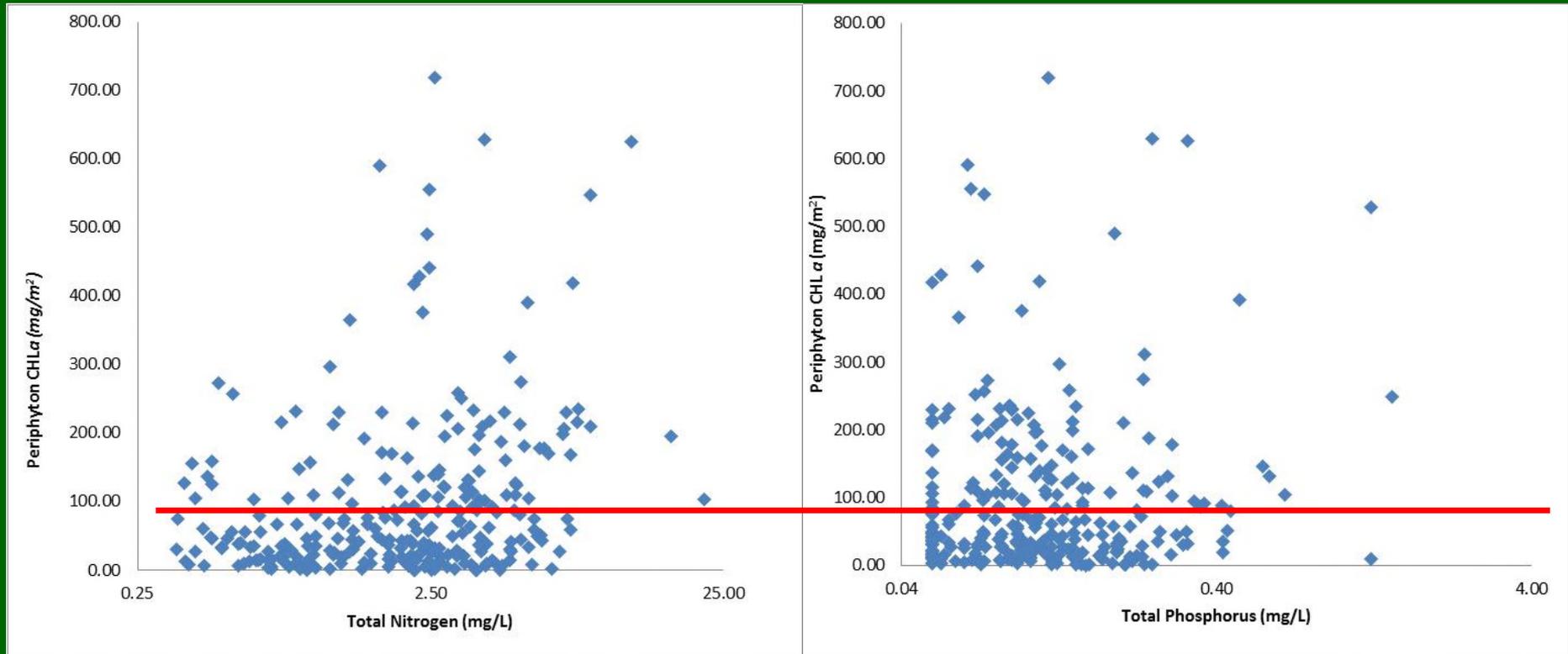
## Presentation Overview

- **IDEM/USGS Nutrient Studies**
- **Indiana's Former Approach**
- **Breakpoint Thresholds**
- **Identifying Reference Sites in Indiana**
- **Major Findings**

# IDEM/USGS Nutrient Studies

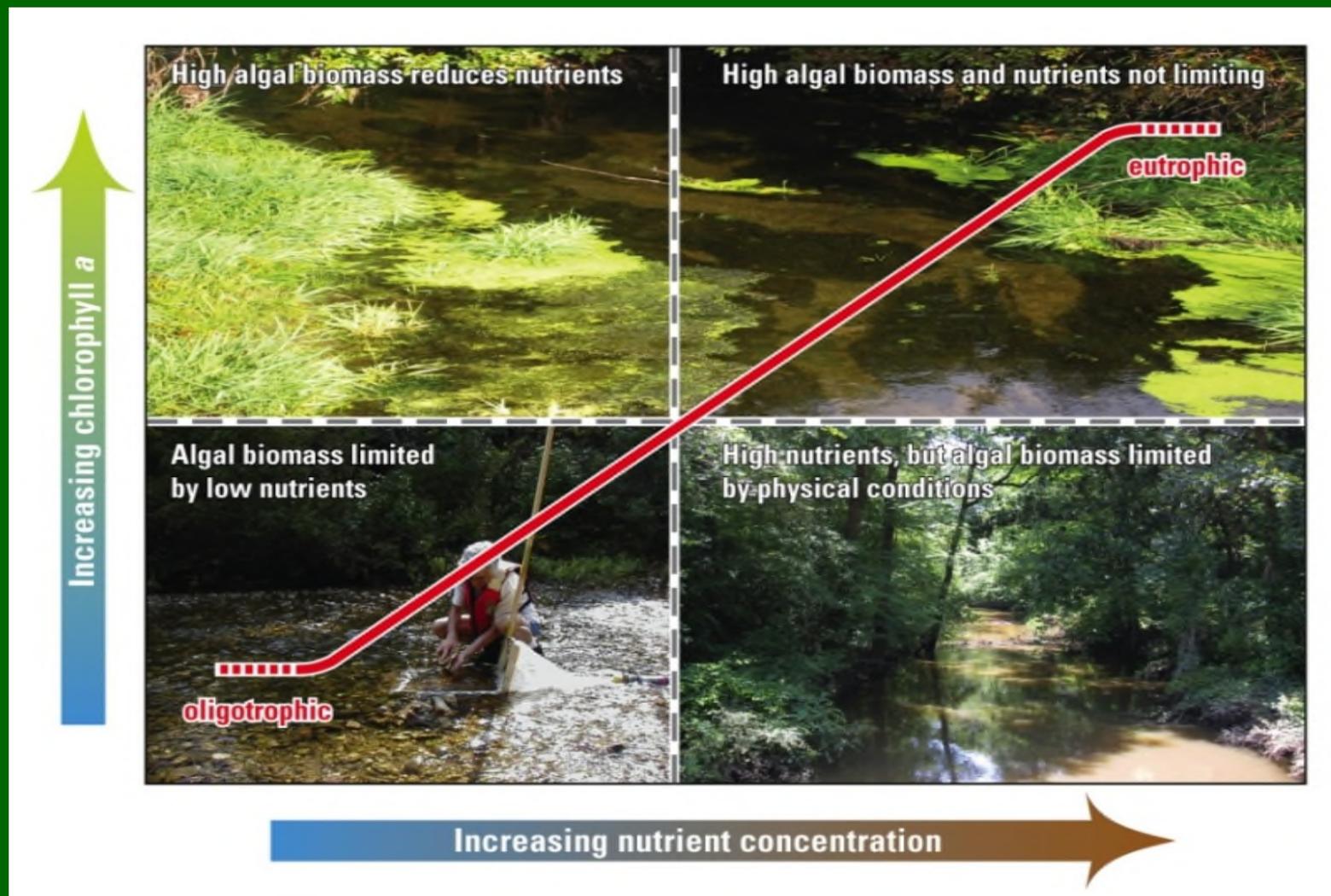
- **2001:** Began collaboration to study nutrients in Indiana rivers and streams
- 10 published USGS-IDEM nutrient reports
  - Initial approaches  
Stressor-response correlation:  
Principal Component Analysis, Spearman Correlations, Correspondence Analysis  
Changes in response variables: LOWESS
  - Current approaches  
Thresholds between stressor and response variables:  
Breakpoint Analysis with Bootstrapping, Quantile Regression  
Differences in biological communities across nutrient gradient:  
ANOSIM and SIMPER

# Initial Approach: Can Nutrients and Algal Biomass, Alone, Determine Nutrient Criteria?



Nutrients and Chlorophyll a  
Concentrations Not Correlated

# Why Relations Between Nutrients and Algal Biomass Are Rarely Found





- **Hypothesis:** Weak to no correlations between nutrients and algal biomass from previous studies might be caused by grouping together of sites with:
  - **Low nutrients and low uptake** (oligotrophic) and
  - **Low nutrients owing to high algal biomass** (nutrient uptake)



## Major Goal of This Study

- Which invertebrate and fish-taxa attributes best reflect the conditions of streams in Indiana along a gradient of nutrient concentrations?
  - Are there ‘oligotrophic’ versus ‘uptake’ sites?
  - If so, is there a species trophic gradient?

# 2005-2009 Study Design

Rotating basin,  
probabilistic

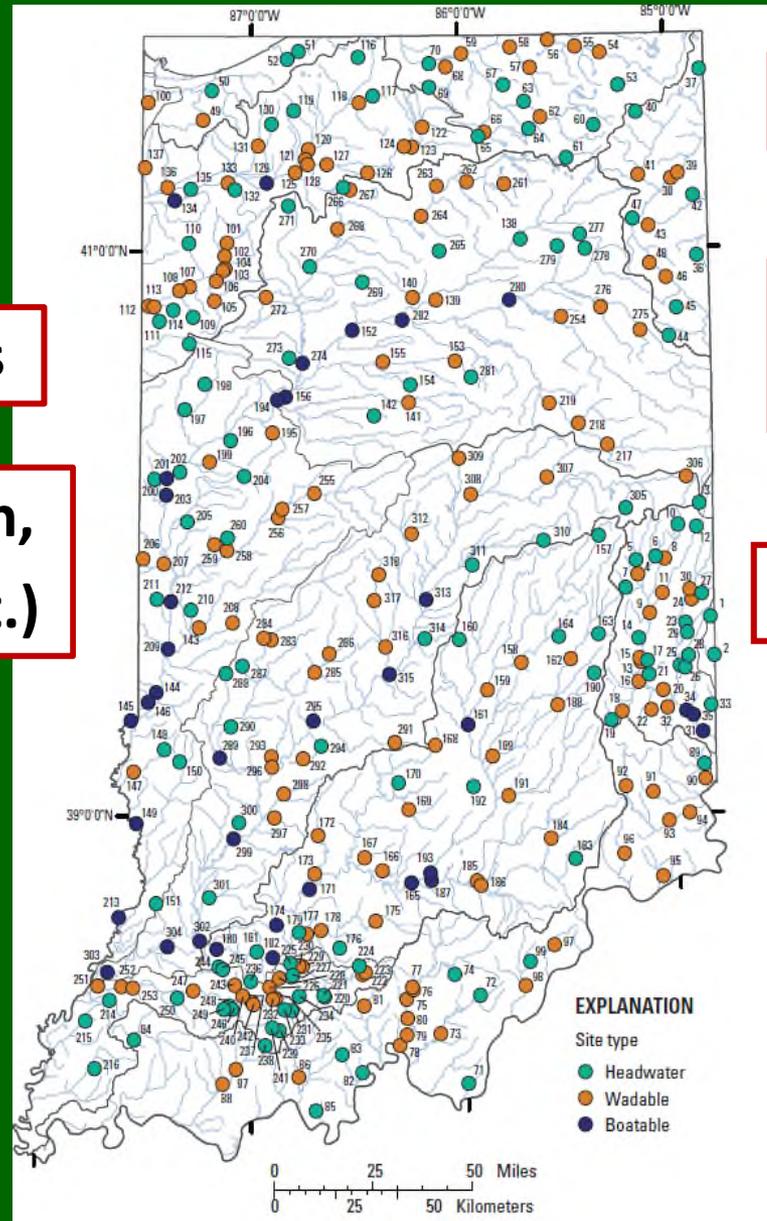
318 sampling sites

34- 38 sites/basin,  
3 times (May-Oct.)

Water chemistry

Fish, inverts,  
habitat

Algal biomass



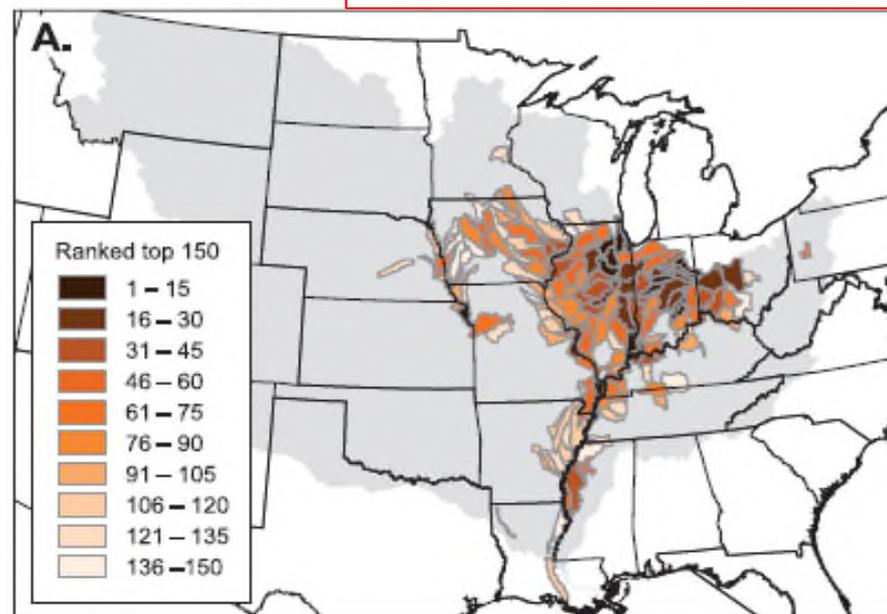
# Where Are the 'Hotspots'?

## Total Nitrogen

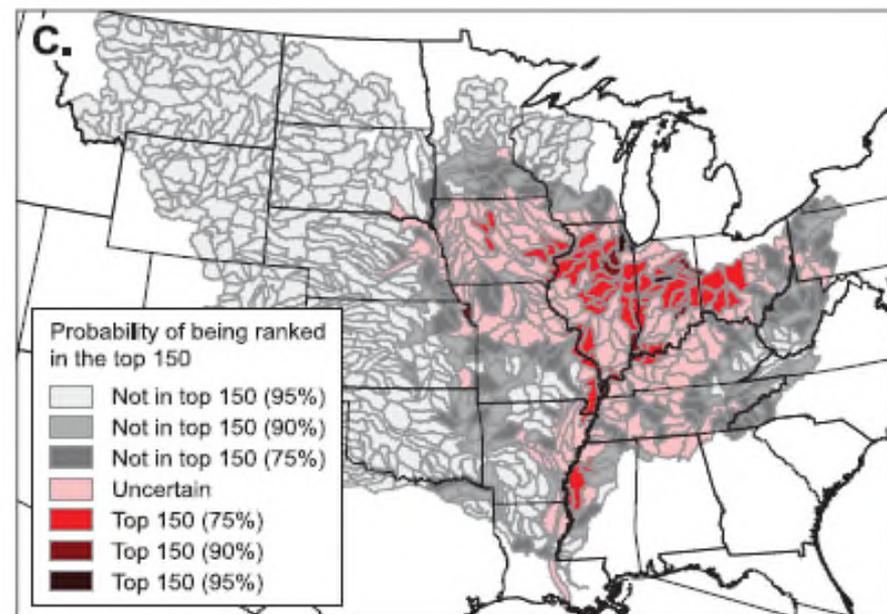
- Cornbelt states dominate
- Indiana has some of the highest ranked hotspots

TOTAL NITROGEN

From: Roberson and others, 2009

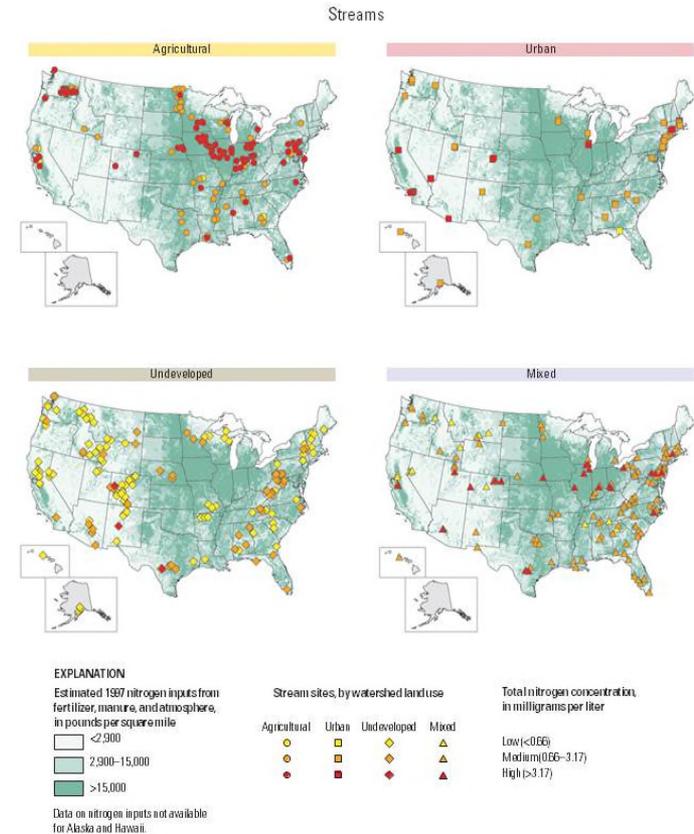


TOTAL NITROGEN





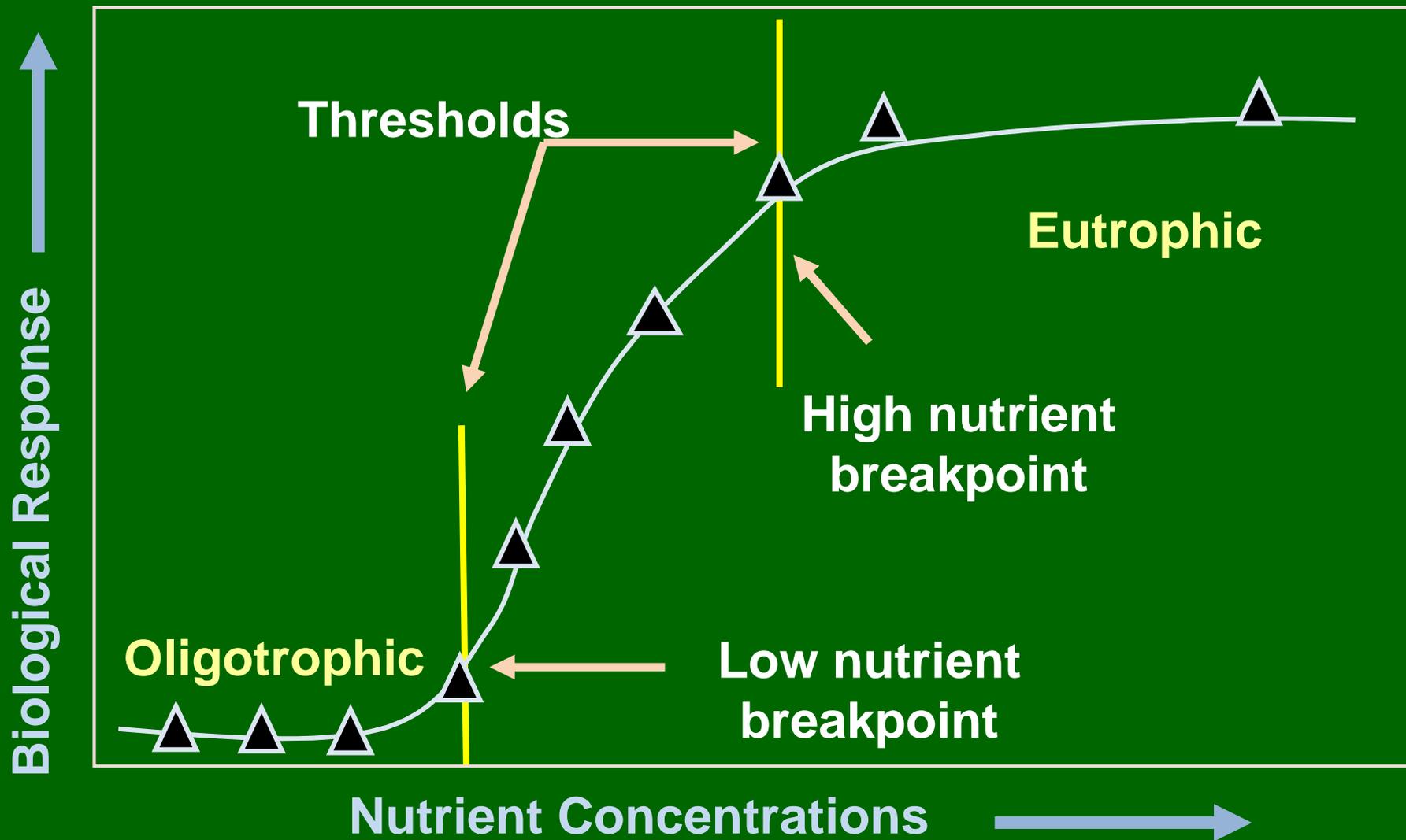
- Indiana and other Cornbelt States have some of the highest instream nutrient concentrations in the country



From Dubrovsky, et al, 2012

# A Conceptual Model:

## Positive Biological Response to Nutrients

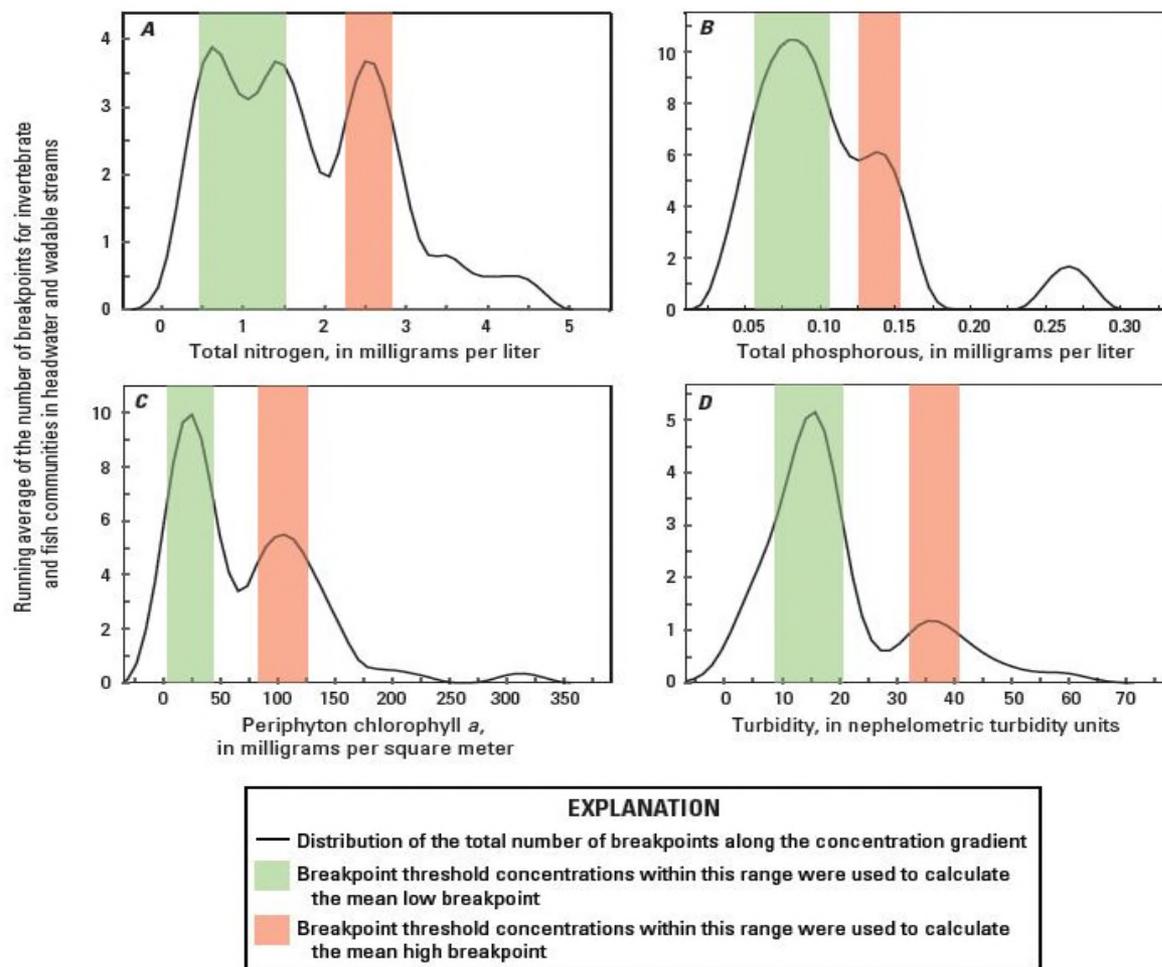


# Identifying Breakpoint Thresholds

- Reduced the list of attributes using statistics and “Best Professional Judgment”
  - Statistically significant
  - Ecologically significant
- List of Relations Included in the Breakpoint Thresholds
  - TN - 29
  - TP - 34
  - Periphyton CHL $\alpha$  - 66
  - Turbidity - 57

# Lower and Upper Breakpoints Using Most Commonly Occurring Breakpoints

- Includes all significant breakpoints from invert and fish measures



**Figure 7.** Gaussian distribution lines of the total number of invertebrate and fish breakpoint threshold concentrations calculated for the headwater and wadable site datasets. *A*, total nitrogen. *B*, total phosphorus. *C*, periphyton chlorophyll *a*. *D*, turbidity. Peaks represent the concentration where the largest influence to biological communities occurred and were used to calculate mean low and mean high breakpoints in the nutrient-enrichment study, 2005–9.



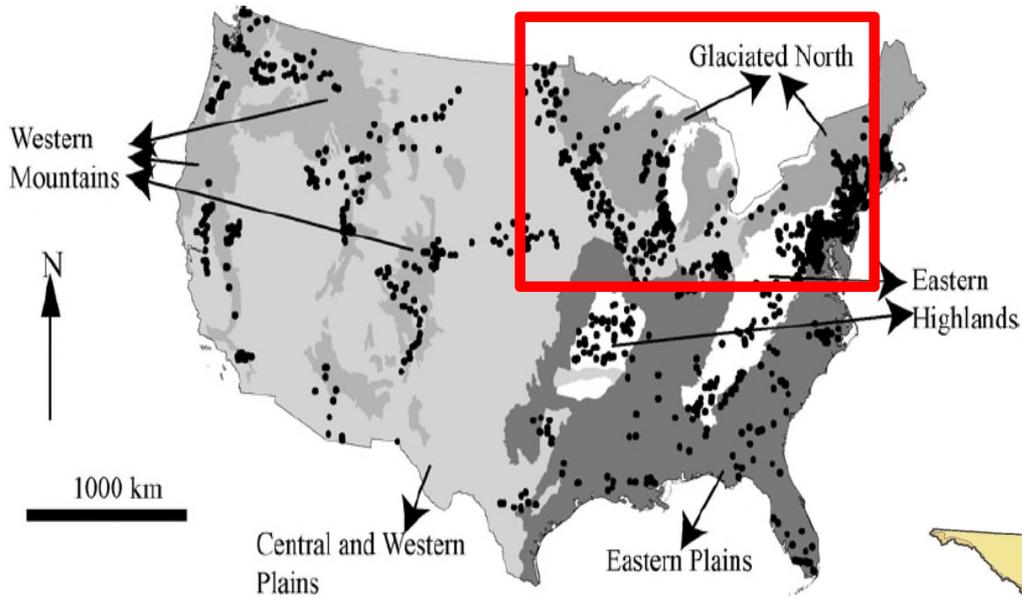
# Biological Breakpoints: Indiana Data

**Table 2.** Mean breakpoint threshold summary for the stressor variables, 2005–9.

[TN, total nitrogen; mg/L, milligrams per liter; N/A, not applicable; TP, total phosphorus; Peri, periphyton; CHL<sub>a</sub>, chlorophyll *a*; mg/m<sup>2</sup>, milligrams per square meter; NTU, nephelometric turbidity units]

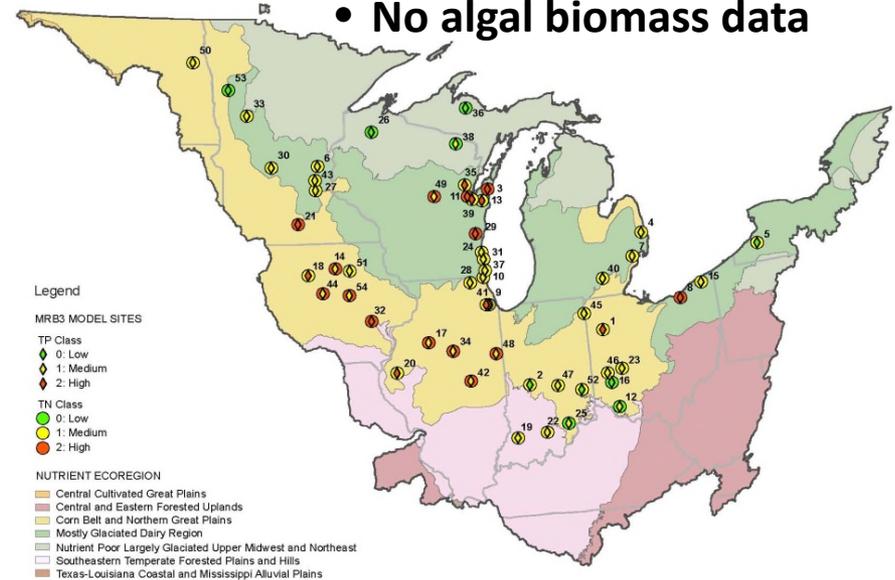
Causal variable	Lowest breakpoint	Highest breakpoint	Mean low breakpoint	Mean high breakpoint	Dodd's oligotrophic-mesotrophic	Dodd's mesotrophic-eutrophic	Biological impact
TN (mg/L)	0.314	4.35	1.03	2.61	0.700	1.50	N/A
TP (mg/L)	.050	.265	.083	.144	.025	.075	N/A
Peri CHL <sub>a</sub> (mg/m <sup>2</sup> )	3.49	312	20.9	98.6	20.0	70.0	N/A
Turbidity (NTU)	3.48	59.1	15.4	36.3	N/A	N/A	50.0

# Midwest Data



## Follows Potapova's Diatom Ecoregions (Potapova and Charles, 2007)

- n = 54
- Biological and nutrient data
- No algal biomass data



# Regional Breakpoint Summary Biological Response

Study	Location	TN (mg/L)		TP (mg/L)	
		Low	High	Low	High
<b>Glaciated North Diatom Ecoregion</b>					
Smith Nutrient IBI (2007)	New York	0.34	1.40	0.018	0.065
Crain and Caskey (2010)	Kentucky wadable	--	--	0.032	--
Miltner (2010)	Ohio	--	--	0.038	--
Heiskary et al (2010)	Minnesota (North and Northwest)	--	--	0.040	0.070
Robertson et al (2008)	Wisconsin (large rivers – inverts)	0.53	1.99	0.040	0.150
Robertson et al (2006)	Wisconsin (wadable streams – fish)	0.54	--	0.055	0.067
Frey et al (2011) wadable	Glacial North (MN, WI, MI)	0.60	1.20	0.030	0.100
Wang et al (2007)	Wisconsin	0.60	--	--	--
Miltner and Rankin (1998)	Ohio	0.61	1.65	0.060	0.170
Robertson et al (2006)	Wisconsin (wadable streams - inverts)	0.61	1.11	0.040	0.067
Robertson et al (2008)	Wisconsin (large rivers) fish	0.63	1.97	0.035	0.150
<b>Plains Diatom Ecoregion</b>					
Caskey et al (2010)	Indiana wadable	2.40	3.30	0.042	0.129
Heiskary et al (2010)	Minnesota (south)	1.77	3.60		
Frey et al (2011)	Central and Western Plains (IL, IN, OH)	1.70	3.50	0.075	0.133
<b>Background nutrient concentrations or trophic levels</b>					
Dodds et al (1998)	National, 33rd and 66th percentiles	0.70	1.70	0.025	0.075
Robertson et al (2006)	Wisconsin (median reference) wadable	0.61	1.10	0.035	--
Robertson et al (2008)	Wisconsin (median reference) large rivers	0.40	0.70	0.035	--

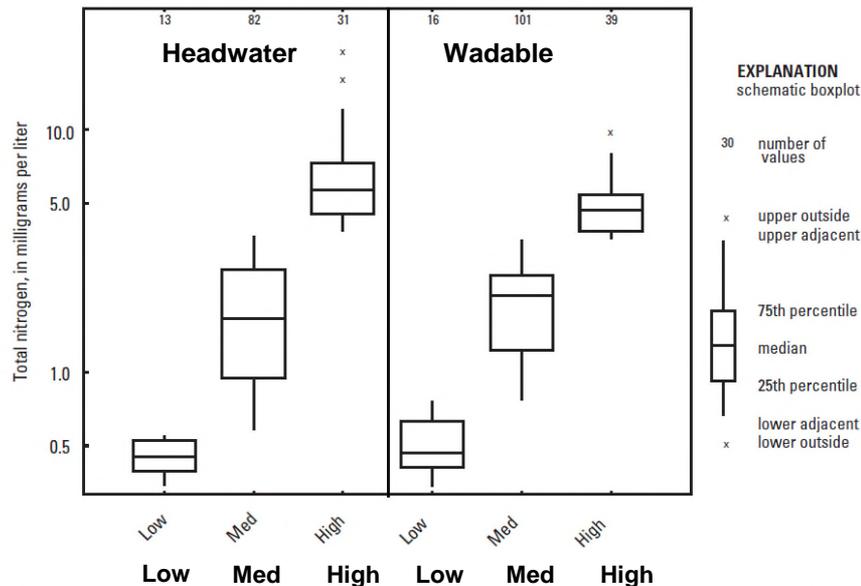


# Defining Nutrient Reference Sites

- **Why are nutrient reference sites important?**
  - Baseline needed to determine nutrient conditions
- **Can we determine nutrient reference sites using nutrient concentrations alone?**
  - Is there a nutrient gradient?
  - Are there biological differences between defined nutrient groups?

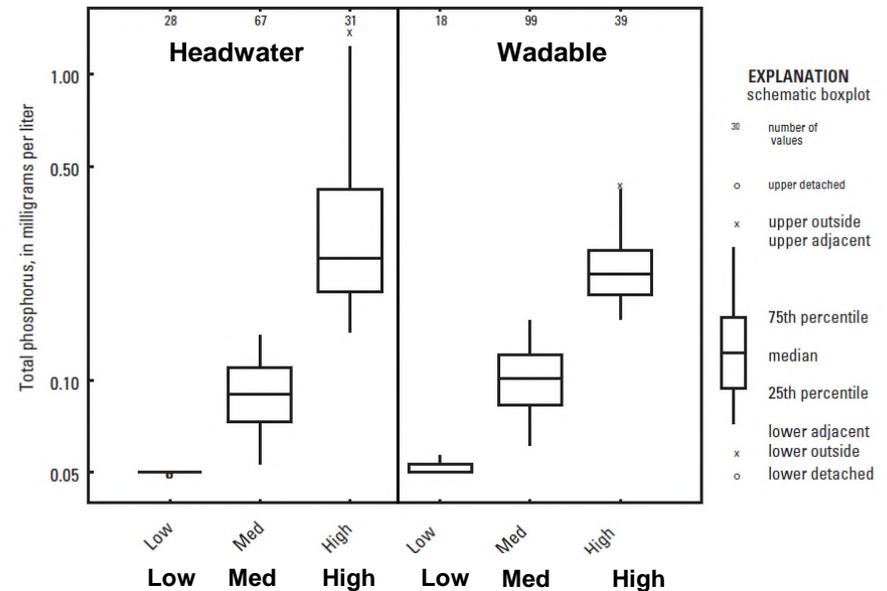
# Nutrient Groups Based on 10<sup>th</sup> and 75<sup>th</sup> Percentiles

## Total Nitrogen



**Figure 3.** Log transformed distribution of mean total nitrogen concentrations in three nutrient groups (Low, Medium, and High) based on nutrient distribution for the selected data sets in the nutrient-enrichment study, 2005–9.

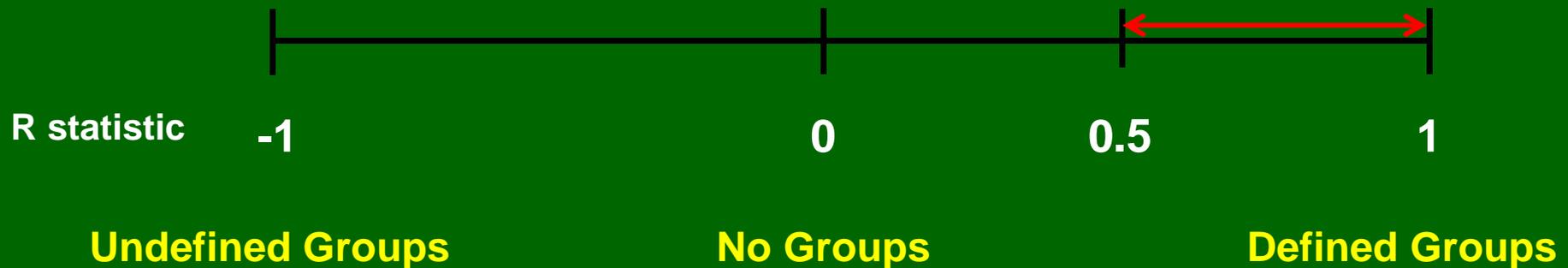
## Total Phosphorus



**Figure 4.** Log transformed distribution of mean total phosphorus concentrations in three nutrient groups (Low, Medium, and High) based on nutrient distribution for the selected data sets in the nutrient-enrichment study, 2005–9.

# Are There Biological Differences Between Defined Nutrient Groups?

- Tests for similarities within defined groups
  - Low, Medium, and High nutrient groups
- Generates  $p$  value and R statistic



Significance defined by  $p$ -value

Analysis of Similarity (ANOSIM)

# No Biological Differences Between Defined Nutrient Groups

- Some results significant
- All R statistics below 0.5
- **Strongest relations:** between Low and High Categories

**Table 4.** Analysis of Similarity (ANOSIM) results for pairwise tests of invertebrate and fish communities in Low, Medium, and High total nitrogen and total phosphorus groups for headwater and wadable sites, 2005–9.

[vs., versus]

Site type	Group comparisons	Invertebrate		Fish	
		R statistics	p-value	R statistics	p-value
Total nitrogen					
Headwater	Low vs. Medium	-0.001	0.488	0.070	0.164
	Medium vs. High	-0.056	.936	-0.020	.652
	Low vs. High	.222	.005	.126	.079
Wadable	Low vs. Medium	.007	.481	-0.003	.490
	Medium vs. High	-0.070	.099	.009	.383
	Low vs. High	.291	.001	.034	.282
Total phosphorus					
Headwater	Low vs. Medium	0.029	0.237	-0.027	0.675
	Medium vs. High	.000	.473	.000	.449
	Low vs. High	.097	.002	.027	.100
Wadable	Low vs. Medium	.101	.099	-0.069	.838
	Medium vs. High	.010	.382	.006	.404
	Low vs. High	.241	.001	.091	.087

# No Biological Gradient Based on Nutrient Concentrations



Central Stonerollers



Caddisfly

Freshwater Snail

Low

Medium

High

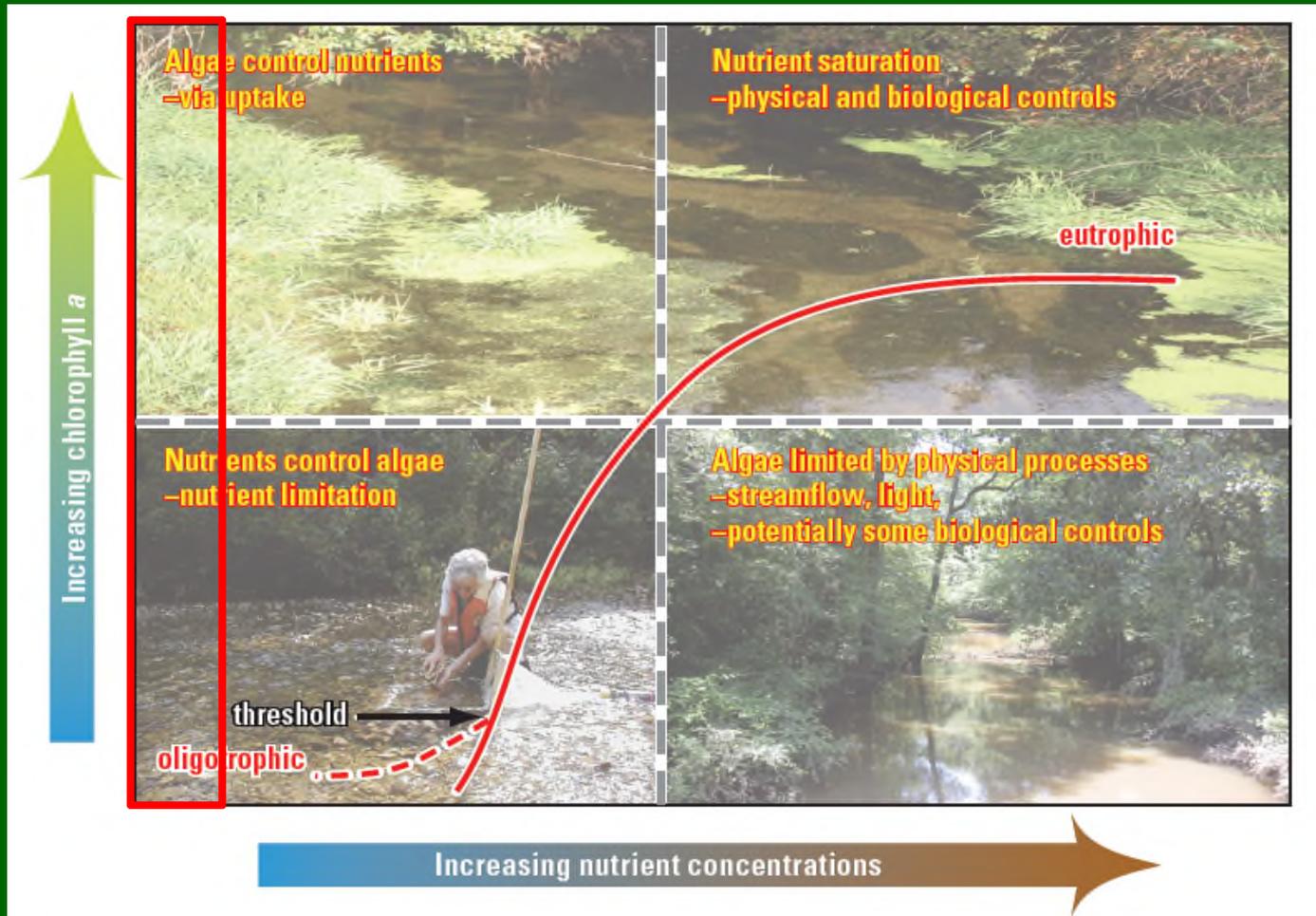
Increasing nutrient concentrations



# Can Algal Biomass Identify Nutrient Reference Sites?

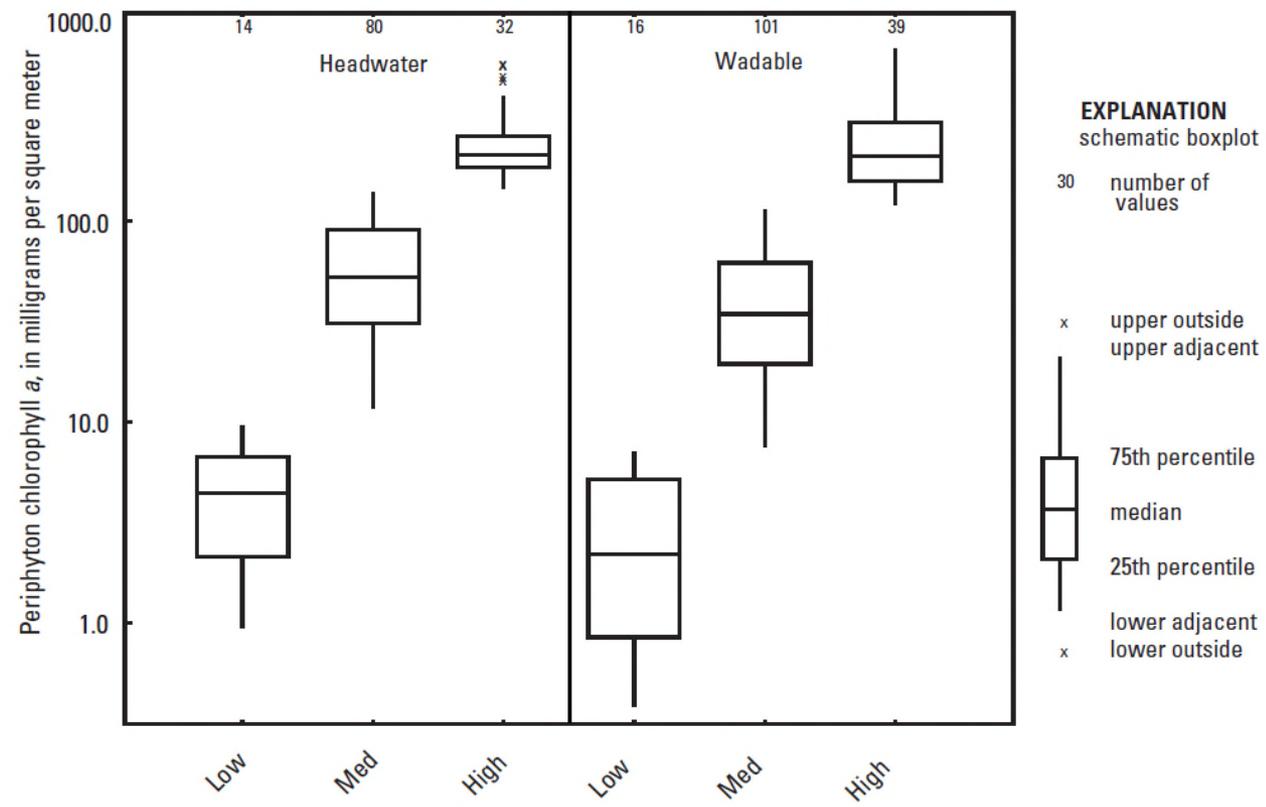
- **No difference in biological communities between sites based on nutrient concentrations**
  - Why would sites with low nutrients and sites with high nutrients have the same communities?
  - **Hypothesis:** Algae use nutrients in the water (nutrient uptake) leading to decreased TN and TP concentrations and increased CHL *a* concentrations leading to community changes

# What Could Explain Similarities in Biology Along Nutrient Gradient?



**Figure 1.** Nutrient-algal biomass conceptual diagram illustrating the relations between nutrient concentrations and algal biomass.

# Periphyton Chlorophyll *a* Concentrations Based on 10<sup>th</sup> and 75<sup>th</sup> Percentiles



**Figure 5.** Log transformed distribution of mean periphyton chlorophyll *a* concentrations in three nutrient groups (Low, Medium, and High) based on nutrient distribution for the selected data sets in the nutrient-enrichment study, 2005–9.



## Defining Nutrient Reference Sites

- **Why are nutrient reference sites important?**
  - Baseline needed to determine nutrient conditions
- **Can we determine nutrient reference sites using nutrient concentrations alone?**
  - Is there a nutrient gradient? **YES**
  - Are there biological differences based on defined nutrient groups? **NO**

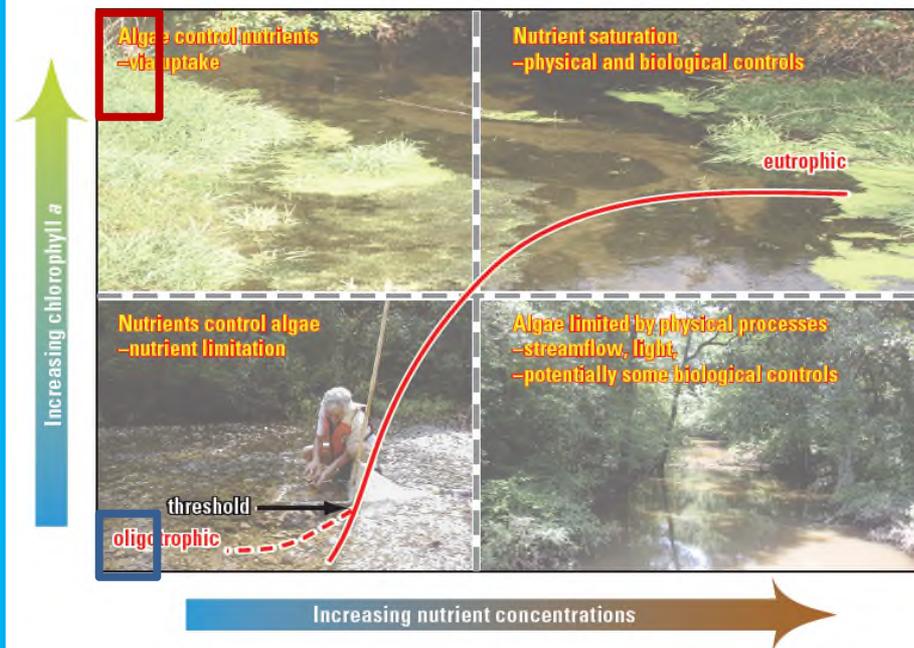
**Nutrient concentrations alone can not be used to define nutrient reference sites**

# Biological Communities Now Different

**Table 6.** Analysis of Similarity (ANOSIM) results for pairwise tests of invertebrate and fish communities based on periphyton chlorophyll *a* categories at low nutrient sites, 2005–9.

[vs., versus; CHL*a*, chlorophyll *a*]

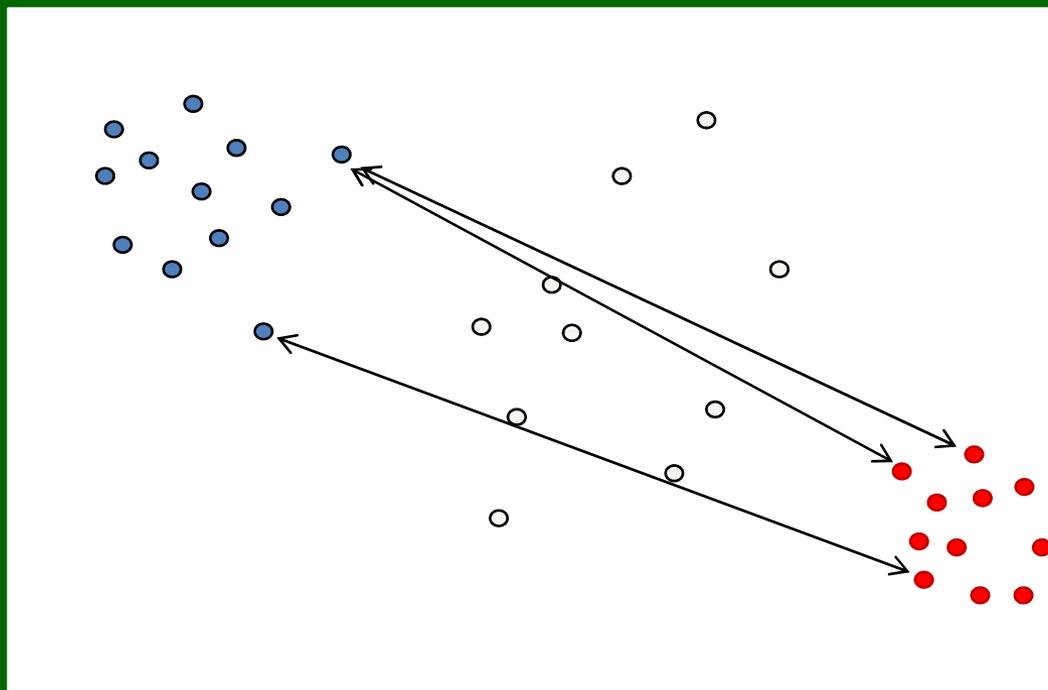
Nutrient group	Invertebrate		Fish	
	R statistic	p-value	R statistic	p-value
Low CHL <i>a</i> vs. High CHL <i>a</i>	0.492	0.001	0.550	0.001
Low CHL <i>a</i> vs. Medium CHL <i>a</i>	.248	.017	.218	.039
High CHL <i>a</i> vs. Medium CHL <i>a</i>	.045	.207	.011	.387



**Figure 1.** Nutrient-algal biomass conceptual diagram illustrating the relations between nutrient concentrations and algal biomass.

# Which Taxa Are Driving Differences Between Communities?

- Identifies taxa that explain the most dissimilarity between groups



Similarity Percentages (SIMPER)

**Table 7.** Average dissimilarity and contributing percentage statistics from Similarity Percentages (SIMPER) analysis for invertebrate and fish communities based on periphyton chlorophyll *a* categories at low nutrient sites, 2005–9.

[CHLa, chlorophyll *a*; sp, species]

Taxa	Average dissimilarity	Contributing percentage	Mean relative percent	
			Low periphyton CHLa category	High periphyton CHLa category
Invertebrate				
<i>Cheumatopsyche</i> sp.	1.39	1.59	4.63	6.59
<i>Physella</i> sp.	1.25	1.44	1.41	6.13
<i>Caenis latipennis</i>	1.16	1.33	.20	3.93
<i>Thienemannimyia</i> sp.	.98	1.12	.60	1.75
<i>Caenis</i> sp.	.97	1.11	.75	1.99
<i>Stalis</i> sp.	.94	1.08	5.48	.81
<i>Dubiraphia minima</i>	.88	1.01	3.00	1.57
<i>Tanytarsus</i> sp.	.85	.98	.15	.95
<i>Simulium</i> sp.	.85	.97	1.72	1.19
<i>Baetis intercalaris</i>	.84	.96	.49	3.20
Fish				
Stonerollers <sup>1</sup>	4.77	6.24	4.09	26.21
Western Blacknose Dace	3.67	4.81	10.36	11.63
Creek Chub	3.59	4.69	6.38	13.39
White Sucker	3.25	4.25	.00	3.68
Striped Shiner	2.86	3.74	.91	5.75
Longear Sunfish	2.76	3.62	19.80	1.86
Mottled Sculpin	2.71	3.55	5.95	4.29
Bluntnose Minnow	2.54	3.33	7.01	4.91
Orangethroat Darter	2.29	2.99	2.93	3.03
Silverjaw Minnow	2.18	2.85	.48	2.14

<sup>1</sup>Represents pooled populations of Central and Largescale Stonerollers with Central Stonerollers comprising 80 percent of the group and Largescale Stonerollers as 20 percent.

- **Contributing percentage:** SIMPER explains how much each taxa contributes to the total dissimilarity between the groups, ordered highest to lowest
- **Mean relative percent** also distinguishes groups

# Major Findings

- **Indiana is a nutrient-enriched region**
  - Mean breakpoints thresholds in this study were 2X greater than areas with less nutrient-enriched streams
- **A nutrient gradient exists**
  - However, biological communities were dominated by nutrient tolerant species
- **Why are we not seeing a biological gradient?**
  - Indiana is nutrient saturated and biological communities have reached equilibrium at these higher nutrient levels

Need to identify nutrient reference sites to determine “oligotrophic” conditions



- **Nutrient concentrations alone may inaccurately assess enrichment in streams**
- **Multiple lines of evidence provide a better understanding of nutrient conditions in streams**
- **Differences in the biological-community composition along an algal biomass gradient in the low nutrient sites may help identify potential oligotrophic versus nutrient uptake sites**
- **Nonpoint source reduction is necessary to reach the desired nutrient levels and biological response**



# Saturated?

<http://in.water.usgs.gov/publications/>

Prepared in cooperation with the Indiana Department of Environmental Management, Division of Water, Assessment Branch

**Breakpoint Analysis and Assessment of Selected Stressor Variables on Benthic Macroinvertebrate and Fish Communities in Indiana Streams: Implications for Developing Nutrient Criteria**

Scientific Investigations Report 2010-5026

U.S. Department of the Interior  
U.S. Geological Survey

National Water-Quality Assessment Program

**Assessment of Nutrient Enrichment by Use of Algal-, Invertebrate-, and Fish-Community Attributes in Wadeable Streams in Ecoregions surrounding the Great Lakes**

Scientific Investigations Report 2011-5009

U.S. Department of the Interior  
U.S. Geological Survey

Prepared in cooperation with the Indiana Department of Environmental Management, Office of Water Quality

**Identifying Nutrient Reference Sites in Nutrient-Enriched Regions: Using Algal, Invertebrate, and Fish-Community Measures to Identify Stressor-Breakpoint Thresholds in Indiana Rivers and Streams, 2005-9**

Scientific Investigations Report 2012-5243

U.S. Department of the Interior  
U.S. Geological Survey