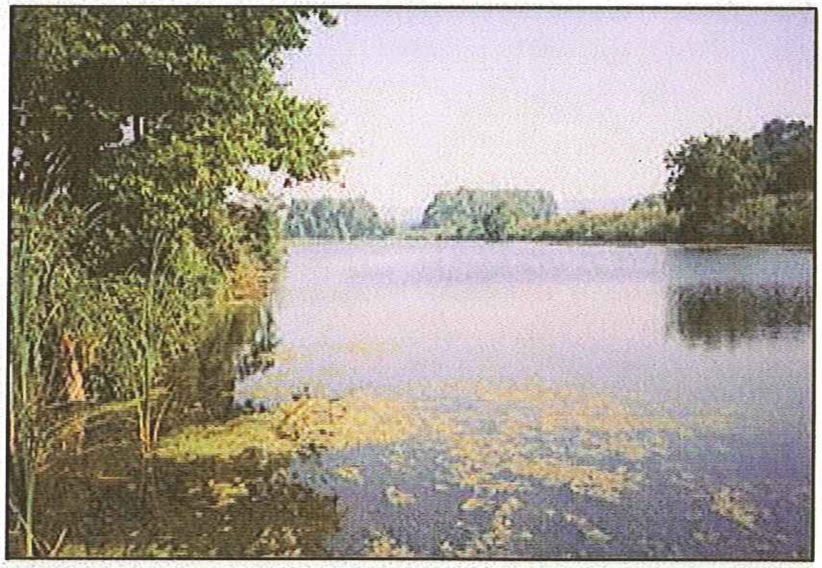
The use of Metaphyton to Evaluate Nutrient Impairment and Proposed Nutrient Criteria for Wetlands and Backwaters in the Upper Mississippi River



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October 2008

Introduction

Anthropogenic enrichment of freshwater systems have largely focused on the impacts of excessive nutrients, primarily nitrogen and phosphorus, in lakes. Targets for improvements in lakes have centered on the control of nuisance sestonic algae, that contribute to algal blooms, algal toxicity problems, foul-tasting drinking water, unsightly conditions and other use impairments. Minnesota has identified nutrient impairment problems in Lake Pepin, a natural tributary delta lake in the Upper Mississippi River system, due to excessive phytoplankton growths during low flow summer conditions (MPCA 2004). The Upper Mississippi River Basin has been the recent focus of watershed-wide nutrient control due to algae-related hypoxic conditions that develop in Gulf Coastal Waters associated with elevated nitrogen and phosphorus inflows from the Mississippi River (US EPA 2008).

The Upper Mississippi River (UMR) bordering Wisconsin contains a highly complex system of aquatic areas besides the main river channel. Aquatic areas include floodplain lakes, lake-like impoundments (above navigation dams), tributary delta lakes, secondary and tertiary stream channels, and contiguous and isolated backwaters along the river's one to two mile wide floodplain corridor (Wilcox 1993). This floodplain community offers a diverse array of aquatic habitat for fish and aquatic life including waterfowl. Annual migrations of hundreds of thousands of waterfowl are attracted to this reach each fall due to the availability of plant foods, aquatic insects and mollusks. Due to relatively low summer suspended solids or turbidity levels, especially in the reach from lower Lake Pepin to Wisconsin's southern border near Dubuque, IA, light penetration is generally sufficient to support large beds of submersed aquatic vegetation (SAV) in many aquatic areas, including portions of the main channel border. SAV is an important biological component in the Mississippi River floodplain ecosystem providing a vital source of food for waterfowl, substrate for invertebrate colonization, refuge for larval and adult fish and a natural barrier to help to reduce sediment resuspension from boats and wind-generated waves (Korschgen 1988 and Janecek 1998).

Beds of SAV in the UMR also support growths of epiphytes including filamentous green algae (Chlorophyta) that often form canopy-like growth over the macrophyte beds during the summer period. Under extreme conditions, carpet-like mats of these filamentous algae rise to the surface during sunny weather being buoyed by oxygen produced through photosynthesis (cover photo). Thick beds of SAV may also harbor duckweeds at the water's surface as a result of the quiescent areas that develop within these expansive aquatic macrophyte communities. Although some level of periphyton production and duckweed development within SAV beds may be considered natural, there is concern that prolonged shading of SAV by metaphyton may seriously threaten the health or composition of the submersed macrophyte community due to reduced growth including reproductive propagule development. In the Upper Mississippi River, there is a particular concern with the underwater light conditions supporting wildcelery (*Vallinseria americana*), which is a ecologically significant member of the SAV community (Korschgen et al. 1997, Kimber et al. 1995a and Kimber et al. 1995b, UMRCC 2003).

Eutrophication research initially theorized that a "switch" from a SAV dominated aquatic system to one dominated by phytoplankton (change in stable states) was mediated by the shading effects induced by sestonic algae. However, it is now generally recognized that nutrient-stimulated epiphytic and filamentous algae coverings of SAV leaves are more likely the primary agent mediating this reduction in light energy and ultimately to the collapse or decline in the SAV plant communities (Phillips et al. 1978, McDougal et al. 1997, Morris et al. 2003 and Hilton et al. 2006). Reductions of SAV communities may occur gradually or catastrophically and can be influenced by other cofounding factors such as excessive turbidity or phytoplankton (Kemp et al. 1983, UMRCC 2003) or water column anoxia that may develop under thick mats of floating plants in nutrient-enriched environments (Morris et al. 2003). The UMR bordering Wisconsin experienced a rapid decline in SAV, particularly wildcelery, during the late 1980s drought though the specific mechanism for the collapse was never determined (Kimber et al. 1995a, Rogers et al. 1995 and McFarland and Rogers 1998). The SAV community in the river reach recovered during the mid-1990s likely in response to improved light penetration (UMRCC 2003).

Advancement in the understanding of nutrient-related impacts on SAV communities strongly suggest that epiphytic filamentous algae, particularly the development of thick metaphyton mats, maybe a key indicator that threatens to SAV health in riverine systems (Hilton et al. 2006). The identification of nutrient conditions contributing to excessive metaphyton cover and biomass need to be identified in order to formulate water quality management or habitat restoration strategies to abate these impacts. This study was focused on defining nutrient concentrations that contribute to nuisance growths of metaphyton in backwater wetland communities in the UMR bordering Wisconsin. This information will provide support for nutrient criteria development and provide initial targets for minimizing the effect of excessive nutrient enrichment in these aquatic systems.

Methods

Mid-summer water chemistry and metaphyton biomass measurements were collected in eleven backwater areas from Wisconsin's Mississippi River floodplain during 2003 to 2007 (Table 1). The sampling sites extended from Pool 5 below Alma, WI to Pool 9 near Desoto, WI (Figure 1). The sites were not selected at random but were chosen to reflect a broad range of metaphyton (filamentous algae and duckweeds) cover and density as well as varying water quality conditions (Figure 2). The selected backwaters represented wetland communities dominated by SAV with minimal areas of unvegetated open water. The backwaters were mostly isolated from major inflows and would only expected to be connected hydraulically to the Mississippi River or nearby tributary streams during periods of high flows or stages. Some sampling locations were sited in areas expected to be strongly influenced by stormwater or municipal wastewater discharges and others were believed to be minimally impacted by point or nonpoint source pollution. A description of each sampling site is included in Table 1.

The spatial extent of surficial metaphyton cover was estimated visually using 2002 and 2005 true-color aerial photographs along with on-site evaluations. Metaphyton cover was assigned a class value of 1 (< 20%) to 5 (>80%) with 20% increments between the five classes (Table 1). Sites where metaphyton cover was sparse or absent (≤ 20%) were identified as reference sites for the purpose of this study.

Field surveys were conducted in late July and early August to obtain measurements during periods of peak metaphyton biomass and associated water quality data. Specific sampling locations were recorded within +/- 9 meters using a Trimble GeoXM GPS receiver. A quarter-meter square floating sampling frame consisting of 2.5 cm OD PVC tubing was placed over areas with surficial metaphyton cover then all floating metaphyton was collected within the frame using a fine-meshed stainless steel strainer (~0.5 mm). Metaphyton was placed in plastic bags and stored in a cooler with ice until processed in the lab. The dominant taxa present in each sample was recorded based on visual inspection. Metaphyton samples were dried at 80 °C in plant dryers to determine dry weight biomass measurements. Plant tissue analysis of plant samples were determined by the University of Wisconsin Soil and Plant Analysis Laboratory in Madison, Wisconsin.

Surface water quality measurements of dissolved oxygen (DO), temperature, specific conductance and pH were collected at approximately 15 cm below the water surface using a YSI 556 multi-parameter meter. The instrument was calibrated following manufacturers instructions prior to each survey. Water samples were collected at a similar depth for nutrient, chlorophyll, turbidity and total suspended solids analysis. In areas with extensive duckweeds, especially *Wolffia*, water samples were filtered in the field with the stainless steel screen (~0.5mm) to remove large plant material. Turbidity measurements were determined using a Hach 2000P turbidity meter. All other measurements of water quality were performed by the Wisconsin State Laboratory of Hygiene in Madison, Wisconsin using EPA-approved procedures. Water quality and metaphyton samples were normally collected by wading in the near-shore littoral area of each backwater sampled. The exception was Stoddard Islands backwater and the Brice Prairie Channel where sampling occurred from a boat in a central location.

Basic statistics, Spearman rank correlations and Kruskal-Wallis one-way AOV, a non-parametric procedure used to evaluate differences between groupings, were derived using Statistix 8 (Analytical Software, 2003).

Results and Discussion

Study Sites

Study sites were separated into two general metaphyton cover groupings (Group 1 $\leq$ 20% and Group 2 >20%) to evaluate general differences in water quality between areas with little versus moderate to abundant metaphyton cover. Metaphyton consisted of filamentous green algae (Chlorophytes) and duckweeds or "lemnids" (*Lemna sp.* and *Wolffia sp.).* The filamentous algae, when present, were usually found covering SAV in a thin blanket or formed mats at the water's surface. The duckweeds were normally floating in clumps or present in extensive mats when they were abundant (Stoddard wetland, Figure 2).

Group 1 sites included three isolated backwaters that had less than less than 20% metaphyton cover and were believed to be minimally impacted by point or nonpoint source nutrient inputs. Due to the sparse presence of metaphyton, Group 1 sites were designated as reference sites since they were presumed to be least impacted by nutrients. These sites included Lizzy Pauls Pond (Pool 5), an isolated wetland south of Coon Creek (Pool 8), and Desoto Pond (Pool 9), an isolated backwater that is influenced by an artesian well (Table 1). Duckweeds were normally absent or present at very low densities at these three sites. Filamentous algae were occasionally observed on SAV at these sites, but their density and spatial coverage were low. Metaphyton biomass at Group 1 sites averaged 3.6 g/m^2; however, the median was 0 g/m^2 since most sites did not contain these plants (Table 2).

SAV was common at all Group 1 sites and was typically dominated by *Ceratophyllum* or unidentified *Potamogetons.* Moderate *Nymphyea* and sparse stands of *Zizania* were scattered along the margins at Lizzy Pauls Pond (Figure 1), a wetland that is relatively free from point or nonpoint source pollutant inputs, located in the backwaters of Pool 5 below Alma, Wisconsin. Water clarity was usually high at the three reference sites due to low suspended particulate matter (low turbidity) or sestonic algae (low chlorophyll *a*), (Table 2). Water depths at reference sites ranged from 0.5 to 0.9 m.

Group 2 sites were moderately to heavily covered with metaphyton (>20 to 100%) and frequently formed thick extensive mats at the surface at the margins of the backwaters where it was abundant. Some of the heaviest growths were encountered in an isolated wetland south of Stoddard, Wisconsin (Figure 2). The north end of this wetland received effluent from the village of Stoddard's wastewater treatment plant through the fall of 2005. *Lemna* and *Wolffia* frequently occurred together at many of the Group 2 sites and it was sometimes difficult to determine which genus was dominant since it was not possible to separate these lemnids when performing biomass measurements. The dominant duckweed genera recorded was based on visual examination of the duckweed mass collected during biomass sampling. Metaphyton biomass of Group 2 sites averaged 73.6 g/m^2 dw and had a median of 59.9 g/m^2 dw. The average was positively biased due to excessive metaphyton biomass (>150 g/m^2 dw) at some sites. The average metaphyton biomass at Group 2 sites was about 17 times the levels determined for the references sites (Table 2).

The SAV at Group 2 sites was typically dominated by *Certophyllum demersum, Elodea canadensis and Myriophyllum sp.* Water clarity in Group 2 sites was generally lower than reference sites due to higher particulate organic material and occasionally higher sestonic algae concentrations, which contributed to higher turbidity levels (Table 2). Mean water depth at Group 2 sites was identical to the reference sites (0.8 m), but the range in depths was greater (0.3 to 1.8 m) at Group 2 sites.

Water Quality and Metaphtyon Cover

Water quality measurements differed significantly between the two metaphyton cover groupings for most of the variables evaluated (Table 2). The reference sites had substantially higher DO concentrations and lower levels of nutrients, particularly inorganic nitrogen (dissolved ammonia-N and nitrite+nitrate-N), total N (TN), dissolved-P (soluble reactive P), particulate-P and total-P (TP). Sestonic chlorophyll *a* concentrations associated with phytoplankton or free-floating filamentous algae were also significantly less at the reference sites versus the Group 2 sites with median levels of 9.6 versus 26.0 ug/L, respectively. It is suspected that some of the chlorophyll *a* concentrations derived for Group 2 sites may have been positively biased due to inclusion of tiny *Wolffia* cells that passed through the filtering screen during the collection of water samples.

Median DO concentrations at Group 2 sites were extremely low (0.8 mg/L) and were indicative of substantial hypoxia with high community respiratory demand, likely due to sediment oxygen demand, aquatic plant respiration and decreased surface re-aeration. The heavy canopy of surface metaphyton that was present at these sites likely contributed to reduced re-aeration and lower photosynthetic rates by aquatic macrophytes, periphyton and sestonic algae due reduced light. Light penetration measurements made in Mississippi River backwaters, as part of a separate evaluation by the author, revealed a thick covering of metaphyton (>25 g/m^2 dw) attenuated 70 to 90% of surface light (Figure 3) and was consistent with other studies (McDougal et al. 1997, Dodds et al. 1999 and Brush & Nixon, 2002). A dense covering of metaphyton likely contributed to lower sub-surface water temperatures observed at the Group 2 sites by reflecting solar radiation or by restricting radiant energy penetration.

Lowest DO concentrations were found at sites that were dominated by *Lemna (*Table 3*)*. Mid-summer monitoring conducted by the federal Long Term Resource Monitoring Program (LTRMP) in Pool 8 between 2005 and 2007 indicated mid-day DO measurements decreased with increased *Lemna* cover (Figure 4a). Reduced DO with increases in metaphyton cover were also observed in this study (Figure 4b), but the DO levels were lower and likely reflected sites that were more isolated with minimal mixing with higher oxygenated flowing water. The combination of high community respiration, reduced photosynthesis and re-aeration contributed to waters that were substantially under-saturated with DO even during mid-day conditions.

The DO concentrations at the sediment-water interface were not determined but anoxia was probable given the very low water column DO levels measured at many sites with high metaphyton biomass. Anoxic bottom waters favor increased rates of dissolved P and ammonia N releases from sediments (James et al. 1995, James et al. 2008 and Wetzel, 1983), which would support the growth of metaphyton if these nutrients are not effectively assimilated by SAV, benthic algae or periphyton. Bottom waters that retain oxygen would be expected to have less internal loading of these nutrients and may allow aquatic macrophytes and periphyton to effectively utilized the dissolved forms of N & P preventing excessive metaphyton growth, provided major external nutrient inputs were not important. The DO content of backwaters and wetlands likely plays an important role in influencing the availability of water-column phosphorus and ammonia nitrogen concentrations that help fuel plant production.

The median dissolved silica concentration was noticeably lower at the reference sites (4.9 mg/L) in comparison to Group 2 sites (11.7 mg/L) where metaphyton biomass was greater (Table 2). It is suspected that greater light penetration at the reference sites yielded conditions more favorable for epiphytic and periphytic diatom growth, which would be expected to contribute to greater assimilation of dissolved silica. In contrast, Group 2 sites had moderate to high metaphyton biomass comprised primarily of *Lemna* and or *Wolffia* and likely contributed to reduce growth of diatoms and other attached algae as a result of reduced light availability due to shading by these lemnids.

Spearman rank correlation analysis revealed metaphyton biomass and cover (cover class) were strongly correlated to most of the water quality measurements performed in this study (Table 4). Metaphyton was positively correlated to nutrients, especially total kjeldahl N (TKN), total organic N, TN, particulate P and TP. Fewer significant correlations were observed between the dissolved forms of N and P and metaphyton biomass. This latter response was likely due to relatively low dissolved N and P concentrations present during these surveys as a result of nutrient assimilation by plants and dentrification. Metaphyton was negatively correlated to total N:P which suggest the metaphyton favored waters that were enriched with P. Metaphyton was also negatively correlated to DO and pH reflecting its ability to grow in hypoxic, reduced conditions. This correlation was likely more associated with the lemnids since the DO content of waters dominated by filamentous algae had higher DO concentrations (Table 3).

Water Quality and Metaphyton Cover Type

Significant differences in water quality measurements were associated with the presence and type of metaphyton cover (Table 3). Sites with no metaphyton or moderately low densities of filamentous algae (median 12.4 g/m^2) had the lowest median TN and TP and higher median DO oxygen concentrations. TN and TP were noticeably greater in areas dominated by *Lemna* where median TP and TN concentrations exceeded 0.6 and 3 mg/L, respectively, and median biomass exceeded 90 g/m^2 dw.

Dissolved forms of N and P were normally present at low concentrations where metaphyton was absent or where metaphyton was dominated by filamentous algae or *Wolffia* (Table 3). Median dissolved NOx-N concentrations were very low and usually at or near the laboratory detection limit of 0.019 mg/L. Areas dominated by *Lemna* exhibited relatively high median concentrations of dissolved ammonia-N (0.302 mg/L) and dissolved P (0.112 mg/L) and were nearly anoxic with a median DO of 0.4 mg/L. It is likely that anoxic bottoms waters contributed to increased rates of internal N & P loading from sediments in areas with extensive *Lemna* cover as discussed previously. The Stoddard wetland site (north end, Figure 2), a site that was normally dominated with a thick carpet of *Lemna* (median biomass >100 g/m^2 dw), was also influenced by a municipal wastewater treatment plant inflow that contributed to elevated ammonia-N (median 1.85 mg/L) and dissolved P (median 1.69 mg/L) at this site.

In general, sites dominated by *Lemna* or *Wolffia* contained higher levels of total organic N (TKN – ammonia N), which was normally the dominant form of N present at the study sites (Table 3). These duckweeds were also present at sites that contained relatively high median particulate P concentrations (0.17 to 0.44 mg/L). However, the median N:P ratio was low (5.8 to 8.6) at sites dominated by duckweeds in comparison to areas where metaphyton was comprised of filamentous algae or where it was absent (12.4 or 13.5, respectively). In general, the relatively low water column N:P ratios at all sites suggest a surplus of P relative to N.

Water Quality, Metaphyton and Chlorophyll Correlations

Spearman rank analysis of raw water quality data versus metaphyton biomass or metaphyton cover indicated numerous significant correlations (Table 4). Highest correlations were associated with particulate forms of N and P. Total N showed slightly greater correlations with biomass or cover as compared to total P. In general, the dissolved forms of N or P exhibited smaller or no correlations to metaphyton biomass or cover. Dissolved oxygen and pH were negatively correlated to biomass and cover as was the N:P ratio. The relationships described for metaphtyon biomass and cover also held for sestonic chlorophyll measurements.

Total N and P also showed many significant correlations to other water quality measurements (Table 4). This was to be expected for dissolved and particulate forms of N or P since they comprise a portion of total of these nutrients. However, there were also strong positive correlations between the various forms of N and P suggesting that all these nutrient measurements were influenced by similar factors.

Metaphyton Tissue Analysis

An evaluation of the elemental composition of plant tissue samples revealed significant differences between filamentous algae, *Lemna* and *Wolffia* (Table 5). These differences were noted for seven elements including N, P, C, Ca, Na, B and Mn. A pair- wise comparison between the three metaphyton types indicated that the elemental composition for *Lemna* and *Wolffia* were generally similar. The exception was Mn where the concentration in *Lemna* was similar to filamentous algae rather than *Wolffia* and C, where the C content of *Wolffia* grouped with filamentous algae. However, these latter results for should be treated with caution due to the small sample size for sites dominated by filamentous algae and where C data were available (n=4). The similar elemental composition for the two lemnids seems reasonable since they are in the same plant family and grow in similar aquatic habitats.

Although N, P, and C composition exhibited significant differences between the three types of metaphyton collected, there were no significant differences in the atomic ratio of C:N, C:P or N:P among these metaphyon taxa. However, the ratios were generally greater in the filamentous algae and the failure to show a difference may have again been due to the relatively small sample size for this type of metaphyton. The mean atomic ratios for the lemnids were very similar and likely reflect similar nutritional needs or assimilation in response to ambient nutrient concentrations. The mean N:P atomic ratio for the filamentous algae was almost identical to the Redfied ratio (Redfield 1934) for these elements (16:1) suggesting that neither nutrient was limiting. The N:P ratio for the lemnids was substantially below the Redfield ratio (~10:1) and suggests either N was limiting or plant tissues contained a surplus of P (luxury consumption). The Redfield ratio was originally based on studies of marine algae and it is assumed to provide a useful reference for metaphyton tissue analysis described here though this should be treated with caution. The observed N:P atomic ratio in the lemnids was consistent with the relatively low N:P ratio (concentration basis) found in water where these plants were dominant (Table 3).

Nutrient composition ratios in phytoplankton and metaphyton, when related to tissue C composition, can be utilized to evaluate potential nutrient limitation (Hall and Cox 1995 and USEPA, 2002) . Nutrient enrichment studies in Manitoba wetlands indicated that P:C (ug/mg) <10 and N:C (ug/mg) <75 reflected N deficiency (Murkin et al. 1994). Application of these nutrient to carbon mass ratios to tissue composition measurements made in the present study indicate that P limitation alone was not likely (Figure 5). Metaphyton was more likely to be limited by N or by both N and P. Murkin's expression of nutrient to carbon ratios in plant tissues in a concentration basis is somewhat confusing and does not follow more conventional measurements that are based on atomic ratios.

Hillebrand & Sommer (1999) in their study of periphyton in Florida wetlands, indicated atomic C:N ratios >10 and N:P <13 in plant tissues reflect N limitation. Further, P limitation was associated with atomic C:P ratios >180 and N:P >22. Application of these atomic ratios to plant composition measurement made in the present study again indicate that N was more likely limiting than P (Figure 6 & 7). There were only two metaphyton samples that suggested P limitation. One sample was a filamentous algae sample collected from the Desoto Pond (reference site) and the other was from the Brice Prairie Channel where ambient TP concentrations were low, 0.026 and 0.054 mg/L, respectively. These TP concentrations reflect low mid-summer TP concentrations for backwaters in the Mississippi River based on a comparison to stratified random sampling data for navigation Pools 4 and 8 where median values typically exceeded 0.15 mg/L for the 1993 to 2006 summer sampling periods (USGS 2008).

Mississippi River backwaters typically have low levels of nitrate as a result of denitrification and nutrient assimilation by aquatic plants (Richardson et al. 2004 and James et al. 2008) which would likely contribute to low inorganic water column N concentrations and low tissue N in metaphtyon as described above. Mats of filamentous green algae found growing over beds of SAV in backwaters typically develop a pale yellow color in mid- to late summer (lower cover photo) and may indicate plant tissues limited by N. Tissue analysis of some of these yellow algal mats, as part of a separate study by the author, indicated relatively low TN (<1.2%) high TP (>0.3%) and low N:P atomic ratios (< 9) suggesting N limitation. Further the C:N atomic ratio of these samples averaged 25 (>20 on a mass basis), which greatly exceeded the Redfield ratio. Reports of unhealthy yellow filamentous algae mats have been reported at C:N mass ratios > 18 based on studies of the filamentous green alga, *Hydrodictoyon reticulatum* in New Zealand (Hall and Cox, 1995). Diversion of nitrate-laden river water into backwaters has been proposed as one method for reducing N loading to the Gulf of Mexico (Mitsch et al. 2001 and James et al. 2008). Such plans will need to be studied carefully since the added N may promote or sustain undesirable growths of nitrogen-limited metaphyton which may negatively impact SAV through light limitation (Phillips et al. 1978, McDougal et al. 1997 and Hilton et al. 2006) or contribute to increased hypoxic conditions (Morris et al. 2003) in SAV beds where water exchange with inflowing oxygenated water is low.

Proposed Nitrogen and Phosphorus Criteria

Nitrogen and phosphorus criteria were derived by evaluating the response of metaphtyon and water column nutrients to DO and by relating nutrient concentrations to metaphyton cover and biomass (Table 6). Dissolved oxygen was used as an indication of nutrient enrichment since excessive mats of metaphyton were associated with low DO as described previously. In addition, elevated TN and TP concentrations were also found in hypoxic waters sampled in this study. Finally, DO serves as a useful target for quantifying nutrient impairment in backwaters and wetlands since hypoxic waters contribute to increased releases of soluble P and N from sediments and DO itself is necessary to sustain aquatic life.

Dissolved oxygen was inversely related to metaphyton biomass, TN and TP (Figure 8). The lowest DO found was typically found in areas where metaphyton biomass was dominated by *Wolffia* and especially *Lemna*. Areas where metaphyton was absent typically had DOs exceeding 3 mg/L with the exception of the Lizzy Pauls Pond site in 2003 when DO was 0.6 mg/L. This low DO was associated with moderately high TP (0.144 mg/L) and sestonic chlorophyll (46.8 ug/L).

Linear regression analysis was utilized to estimate TN and TP concentrations that corresponded to various DO target concentrations. To perform this analysis, TN and TP concentrations greater than 2.0 and 0.16 mg/L, respectively, where omitted to concentrate on that region of the DO vs TN or DO vs TP response that appeared be linear and to discard high nutrient concentrations where nutrients were not likely limiting metaphyton growth (Figure 9). The 1 mg/L DO target was arbitrarily selected as the minimum endpoint in attempt to restrict complete mid-day anoxia. The 3 mg/L represented a "breakpoint" in the DO distribution and metaphyton biomass was typically lower at concentrations exceeding this value (Figure 8). A DO target of 5 mg/L was selected based on Wisconsin's DO criterion for fish and aquatic life (Wisconsin Adminstrative Code NR 102.04 (4)). Utilizing the regression models presented in Figure 9, DO targets of 1, 3 and 5 mg/L corresponded to mid-summer average TP concentrations of 0.150, 0.113 and 0.075 mg/L and TN concentrations of 1.86, 1.37 and 0.88 mg/L, respectively (Table 6). Caution needs to be applied when utilizing these DO-based TP and TN criterion since the unexplained variance in the regressions were very high. Nutrient criteria based on DO-nutrient relationships alone are not warranted.

Metaphyton cover was used as a second approach for defining TN and TP criteria. In this case the average and median TN & TP concentrations were determined for sites with <20% and <60% metaphyton cover (Table 6). Sites with greater than 60% cover were not considered since these areas were normally associated with hypoxic conditions (median DO <2 mg/L), (Figure 4b). An estimate of the TN and TP levels associated with a metaphyton cover <40% was derived based on an average of the ≤20% and ≤60% cover groupings due to an absence of sites assigned to the 21-40% cover class. The TP criteria for ≤20% and ≤60% cover groupings were similar to TP criteria based on a target DO for 5 and 1 mg/L, respectively. The TN criteria based on metaphyton cover were generally lower than those based on DO and were relatively insensitive to changes in cover. The TP and TN criteria based on the average of the ≤20 and ≤60% cover groupings were similar to nutrient criteria derived for a target DO of 3 mg/L.

A third approach for defining nutrient criteria was based on defining TP and TN concentrations at different levels of metaphyton biomass. Three biomass targets were considered 0, <10 and <25 g/m^2 dw (Table 6). Areas with no metaphyton reflected sites that were believed to be least influenced by nutrients and had higher DO conditions due to improved light penetration and surface re-aeration. The upper biomass limit of 25 g/m^2 was chosen based on the rapid increase in metaphyton-induced light attenuation at levels exceeding this value (Figure 3). Adequate light penetration is important to support photosynthetic inputs of DO and the growth and development of SAV which provide important habitat for fish and aquatic life and a critical source of food for migrating waterfowl. The metaphyton biomass of 10 g/m^2 reflected areas with relatively low biomass with 92% of the reference site samples falling below this threshold. The TN and TP criteria derived using these three biomass targets were within the same range as those derived using DO and metaphyton cover described above (Table 6).

A tiered approach for protecting Mississippi River backwaters and wetlands from excessive levels of nutrients is suggested. For wetlands that have an important functional value for fish and aquatic life and have been shown to have diverse or unique aquatic plant communities, especially rare or threatened plant species, nutrient criteria that target a mid-summer/mid-day DO concentration of 5 mg/L and ≤20% metaphtyon cover may be desirable. The average TP and TN criteria for these targets are 0.077 and 0.95 mg/L, respectively. For other wetlands and backwaters, it is suggested that the nutrient criteria be targeted on a DO of 3 mg/L and metaphyton biomass <25 g/m^2 dw. The average TP and TN criteria associated with these targets are 0.107 and 1.23 mg/L, respectively.

The proposed nutrient criteria to limit nuisance growths of metaphyton in Mississippi River backwaters and wetlands are generally consistent with previous estimates. Eutrophication modeling studies of duckweeds in Netherlands indicated minimum summer average TP and TN concentrations of 0.19 and 1.3 mg/L, respectively, were predicted to yield 50% duckweed coverage in ditches receiving agricultural drainage (Liere et al. 2007). Murkin (et al. 1994) in their studies of unproductive Manitoba wetlands indicated nutrient additions of 0.03 mg/L TP and 1.6 mg/L TN were too low stimulate wetland productivity. However, they cite the work of Campeau (1990) who found TP and TN concentrations of 0.20 and 2.4 mg/L, respectively, in these same wetlands resulted in a rapid increase in algae levels contributing to oxygen depletion at night.

The second tier summer average TP criterion concentration (0.107 mg/L) proposed in this work matches that recommended in USEPA's "Redbook" (0.1 mg/L) to "prevent plant nuisances in streams or other flowing waters" (USEPA 1977 and Mackenthun, 1973). This concentrations is also almost identical to the proposed TP criterion (0.105 mg/L) recommended for large rivers in Wisconsin (James Baumann, WDNR, Personal Communications) and the proposed TP target (0.1 mg/L) for the Lake Pepin nutrient TMDL (Steven Heiskary, MPCA, Personal Communications).

Summary & Conclusions

Water quality metaphyton samples were collected mid-summer in eleven backwater wetland areas in the Upper Mississippi River bordering Wisconsin during 2003 to 2007. Metaphyton consisted of filamentous green algae and duckweeds (*Lemna* and *Wolffia*). Highest metaphyton cover and biomass were associated with low DO and high TP and TN concentrations. Reference sites where metaphyton was absent or contained less then 20% cover exhibited relatively low TP and TN concentrations and higher DO. Waters containing extensive mats of *Lemna* were nearly anoxic with a median DO of 0.4 mg/L and relatively high dissolved P and ammonia N, 0.11 and 0.30 mg/L, respectively. Anoxic conditions may have contributed to increased rates of internal P and N (ammonia) releases from sediments and likely acted to support nuisance growths of metaphyton, especially in areas receiving external nutrient inputs or in areas where nutrients were not effectively assimilated by other aquatic plants. Water column ratios of TN/TP were typically less than 10 in areas dominated by duckweeds and suggests a surplus of P relative to N.

Plant tissue analysis of metaphyton indicated significant differences in elemental composition between filamentous algae and *Lemna* and *Wolffia*. However, the composition of *Lemna* and *Wolffia* were generally similar and likely reflect their close association (same plant family) and were often found growing together. The ratios of C:N, C:P and N:P between the three types of metaphyton were not significantly different. The ratios C, N & P provided an indication of nutrient limitation. Plant tissue analysis indicated metaphyton was more likely to be limited by N or by both N & P rather than P alone. In two situations where plant tissue analysis suggested P limitation, ambient TP concentrations were relatively low (< 0.06 mg/L). Metaphyton nutrient composition indicating N limitation in backwaters reflects the low ambient water column TN concentrations due to denitrification and inorganic N assimilation by aquatic plants. The introduction of nitrate-laden waters into backwaters mid- to late summer may have an undesirable effect of promoting or sustaining nuisance growths of N-limited metaphyton.

Nutrient criteria for TN and TP were derived using DO, metaphyton cover and biomass as endpoints. DO was identified as a nutrient-related target since it is an important factor influencing internal nutrient loading from sediments and it is necessary for fish and aquatic life. Nutrient criteria derived from DO alone are not recommended due to the high unexplained variance in DO-nutrient regressions. Metaphyton cover and biomass targets were proposed to limit hypoxic conditions and the development of excessive metaphyton mats that seriously reduce light penetration, which may negatively impact SAV. Two nutrient criteria tiers are suggested. For backwater wetland communities that have an important functional value for fish and aquatic life and contain unique plant communities, average summer TP and TN concentrations should not exceed 0.077 and 0.95 mg/L, respectively. For other backwater aquatic areas, average TP and TN should not exceed average summer concentrations of 0.107 and 1.23 mg/L, respectively. The latter TP criterion is very similar to that recently proposed for large rivers in Wisconsin and to reduce eutrophciation problems in Lake Pepin.

Acknowledgements

The author would like to thank Jeff Houser, USGS, William Richardson, USGS, Steve Hesikary, MPCA, Howard Markus, MPCA, Shawn Giblin, WDNR, for their valuable review and comments on an early draft of this manuscript. Jeff Janvrin assisted in the preparation of the site map and also provided useful comments on this work. The author would like to thank USGS and the WDNR field station for providing additional water quality and metaphyton observations during summer field collections in Pool 8 between 2005 and 2007.

References

Analytical Software 2003. Statistix 8. Tallahasse, Florida.

Campeau, S. 1990. The relative importance of algae and vascular plant detritus to freshwater food chains. M. Sc. Thesis, McGill University.

Brush, M.J. and S.W. Nixon. 2002. Direct measurements of light attenuation by epiphytes on eelgrass *Zostera marina*. Mar. Ecol. Prog. Ser. 238:73-79.

Dodds, W.K., B.J.F. Biggs and R.L. Rowe. 1999. Photosynthesis-irradiance patterns in benthic microalgae: Variations as a function of assemblage thickness and community structure. J. Phycol. 35:42-53.

Hall, J.A. and N. Cox 1995. Nutrient concentrations as predictors of nuisance *Hydrodictoyon reticulatum* populations in New Zealand. J. Aquatic Plant Manage. 33:68-74.

Hillebrand, H. and U. Sommer. 1999. The stoichiometry of benthic microalgal growth: Redfield proportions are optimal. Limnol. Oceanogr. 44:440-446.

Hillton, J. M. O'Hare, M.J. Bowes. and J.I. Jones 2006. How green is my river? A new paradigm of eutrophication in rivers. Science of the Total Environment. 365:66-83.

James, W.F., J.W. Barko and H.L.Eakin. 1995. Interval phosphorus loading in Lake Pepin, Upper Mississippi River. J. Freshwater Ecology. 10(3):269-276.

James, W.F., W.B. Richardson and D.M. Soballe. 2008. Contributions of sediment fluxes and transformations to the summer nitrogen budget of an Upper Mississippi River backwater system. Hydrobiologia 598:95-107.

Janecek, J.A. 1998. Fishes interactions with aquatic macrophytes with special reference to the Upper Mississippi River System. Upper Mississippi River Conservation Committee Fisheries Section. Rock Island, IL. 57 pp.

Kimber, A., J.L. Owens, and W.G. Crumpton. 1995a. Light availability and growth of wildcelery (*Vallinseria Americana*) in the Upper Mississippi River backwaters. Regulated Rivers: Research & Management. 11:167-174.

Kimber, A., C.E. Korschgen, and A.G. van der Valk. 1995b. The distribution of *Vallisneria americana* seeds and seedling requirements in the Upper Mississippi River. Can. J. Bot. 73:1966-1973.

Korschgen, C.E., W.L. Green, and K.P. Kenow 1997. Effects of irradiance on growth and winter bud production by *Vallisneria Americana* and consequences to its abundance and distribution. Aquat. Bot. 58:1-9.

Korschgen, C.E. 1988. American wildcelery (*Vallisneria Americana*): Ecological considerations for restoration. U.S. Fish and Wildlife Service, Fish and Wildlife Technical Report 19. 24 pp.

Liere, L. van, J.H., H.P. Janse, and G.H.P. Arts. Setting critical nutrient values for ditches using the eutrophication model PCDitch. Aquatic Ecology. 41:443-449.

Mitsch, W.J., J.W. Day, J.W. Gilliam, P.M. Groffman, D.L. Hey, G.W. Randall, and N.Wang. 2001. Reducing nitrogen loading to the Gulf of Mexico from the Mississippi River Basin: Strategies to counter a persistent ecological problem. Bioscience, 51(5):373-388.

Morris, K. P.C. Bailey, P.I. Boon and L. Hughes. 2003. Alternative stable states in aquatic vegetation of shallow urban lakes. II. Catastrophic loss of aquatic plants consequent to nutrient enrichment. Mar. Freshw. Res. 54:201-215.

McDougal,R.L., L.G. Goldsborough and B.J. Hann. 1997. Responses of prairie wetland to press and pulse additions of inorganic nitrogen and phosphorus: production by plankton and benthic algae. Arch. Hydrobiol. 140(2):145-167.

Mackenthun, K.M. 1973. Toward a cleaner aquatic environment. Environmental Protection Agency, Washington, D.C.

Minnesota Pollution Control Agency 2004. Lake Pepin Watershed TMDL Eutrophication and Turbidity Impairments Project Overview. Water Quality/Impaired Waters #9.01a. St. Paul, MN. (http://www.pca.state.mn.us/publications/wq-iw9-01a.pdf).

Murkin, H.R., J. B. Pollard, M.P. Stainton, J.A. Boughen and R.D. Titman. 1994. Nutrient additions to wetlands in the Interlake region of Manitoba Canada: effects of periodic additions throughout the growing season. Hydrobiologia. 279/280:483-495.

Phillips, G.L., D. Eminson and B. Moss. 1978. A mechanism to account for macrophyte decline in progressively eutrophicated freshwaters. Aquat. Bot. 4:103-126.

Redfield, A.C. 1934. On the proportions of organic derivatives in sea water and their relation to the composition of plankton. James Johnson Memorial Volume. Liverpool: Liverpool University Press. p. 176-92.

Richardson, W.B., E.A. Strauss, L.A. Bartsch, E.M. Monroe, J.C. Cavanaugh, L. Vingum and D. M. Soballe. 2004. Denitrification in the Upper Mississippi River: rates, controls, and contribution to nitrate flux. Can. J. Fish. Aquat. Sci. 61:1102-1112.

Upper Mississippi River Conservation Committee. 2003. Proposed light-related water quality criteria necessary to sustain submersed aquatic vegetation in the Upper Mississippi River. Water Quality Technical Section. 6 pp. (http://www.mississippi-river.com/umrcc/).

U.S. Environmental Protection Agency. 1997. Quality Criteria for Water. Washington, D.C. 501 pp.

U.S. Environmental Protection Agency 2002. Methods for evaluating wetland condition. #16 Vegetation-based indicators of wetland nutrient enrichment. Office of Water, Washington, DC. EPA-822-R-02-024. 22 pp.

U.S. Environmental Protection Agency 2008. Mississippi River Basin and Gulf of Mexico Hypoxia. Office of Wetlands, Oceans and Watersheds. Washington, DC. (http://www.epa.gov/msbasin/).

U.S. Geological Survey. 2008. Graphical Water Quality Database Browser – Stratified Random Sampling, Upper Midwest Environmental Sciences Center, La Crosse, WI. (http://www.umesc.usgs.gov/ltrmp.html).

Vollenwider, R.A. 1968. Scientific fundamentals of eutrophication of lakes and flowing waters, with particular reference to nitrogen and phosphorus as factors in eutrophication. OECD. Paris, France.

Wilcox, D.B. 1993. An aquatic habitat classification system for the Upper Mississippi River system. US Fish Wildlife Service. Tech. Rep. 93-T003.

Wetzel, R.G. 1983. Limnology. Sec. Ed. Saunders College Publishing. Philadelphia, Pa. 767 pp.

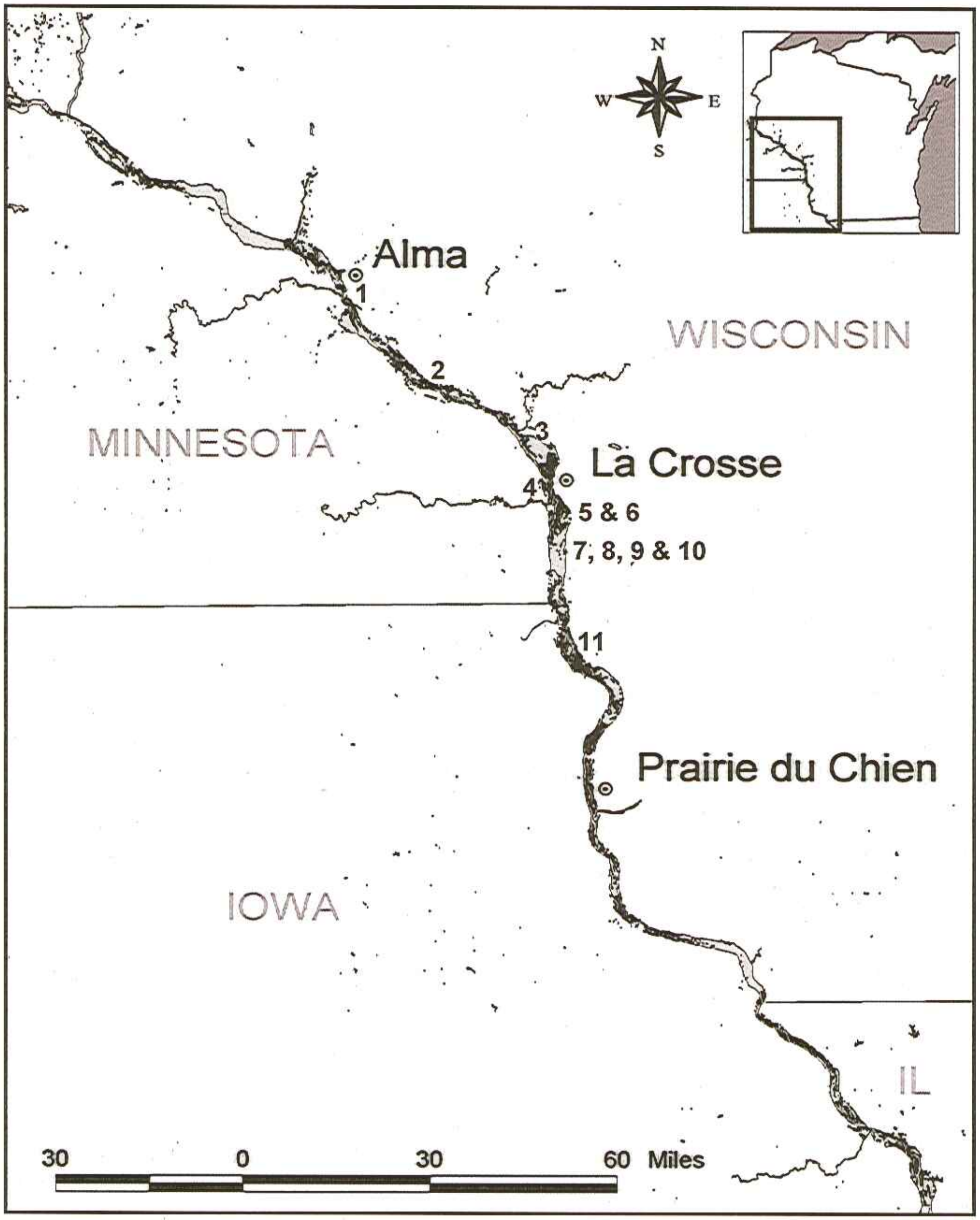


Figure 1. Upper Mississippi River study sites*

No.	Site Description
1	Lizzy Pauls Pond, Pool
2	Lock & Dam 5a wetland, Pool 5a
3	Brice Prairie Channel, Pool 7
4	Pettibone Park wetland, Pool 8
5	Goose Island Access Rd wetland, Pool 8
6	Goose Island Road wetland near first road culvert, Pool 8
7	Stoddard Islands backwater, Pool 8
8	Stoddard wetland N. end, Pool 8
9	Stoddard wetland S. end, Pool 8
10	Wetland S. of Coon Creek, Pool 8
11	Desoto Pond, Pool 9

*detailed site maps provided on following pages

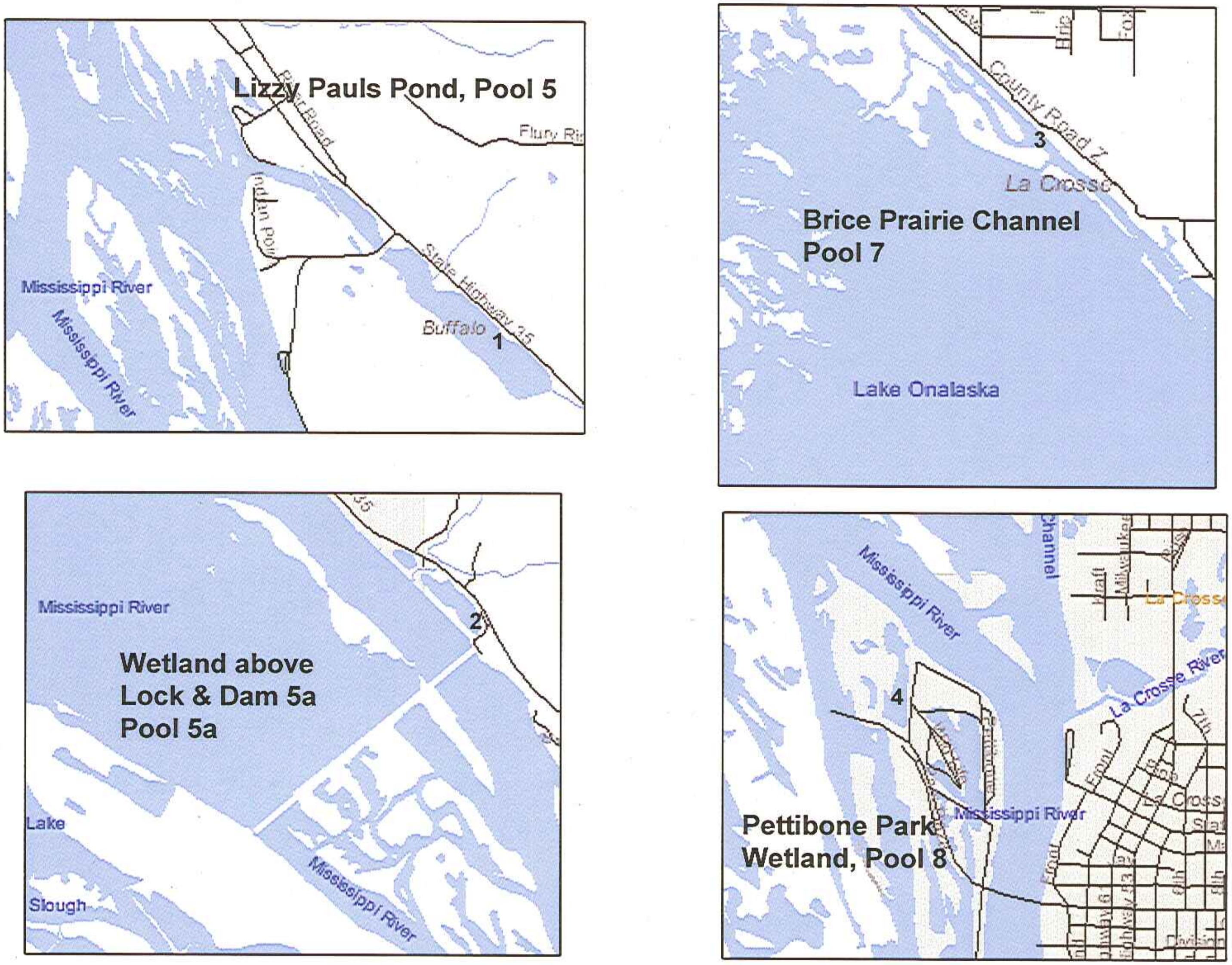


Figure 1. Continued – Detailed site maps.

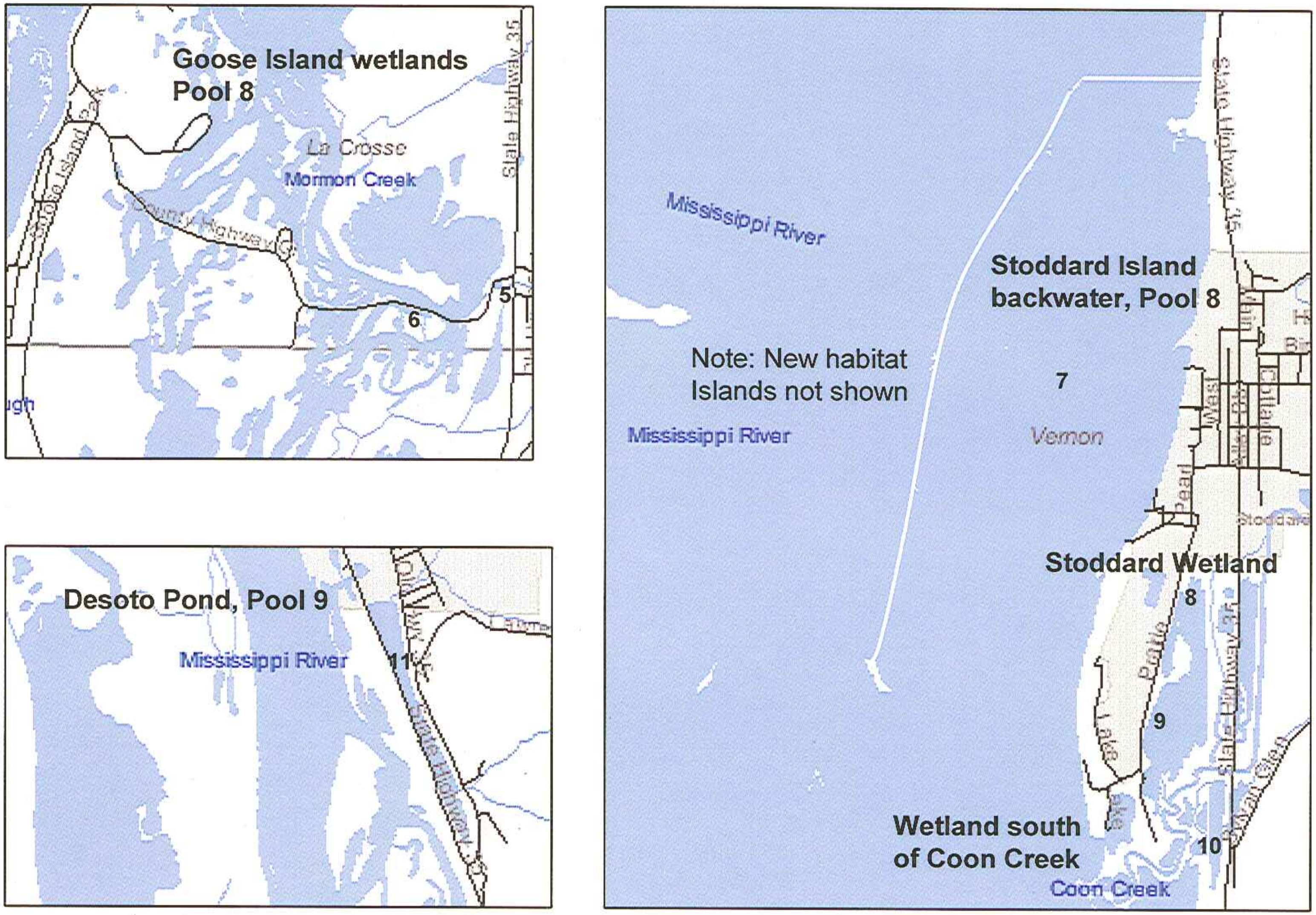
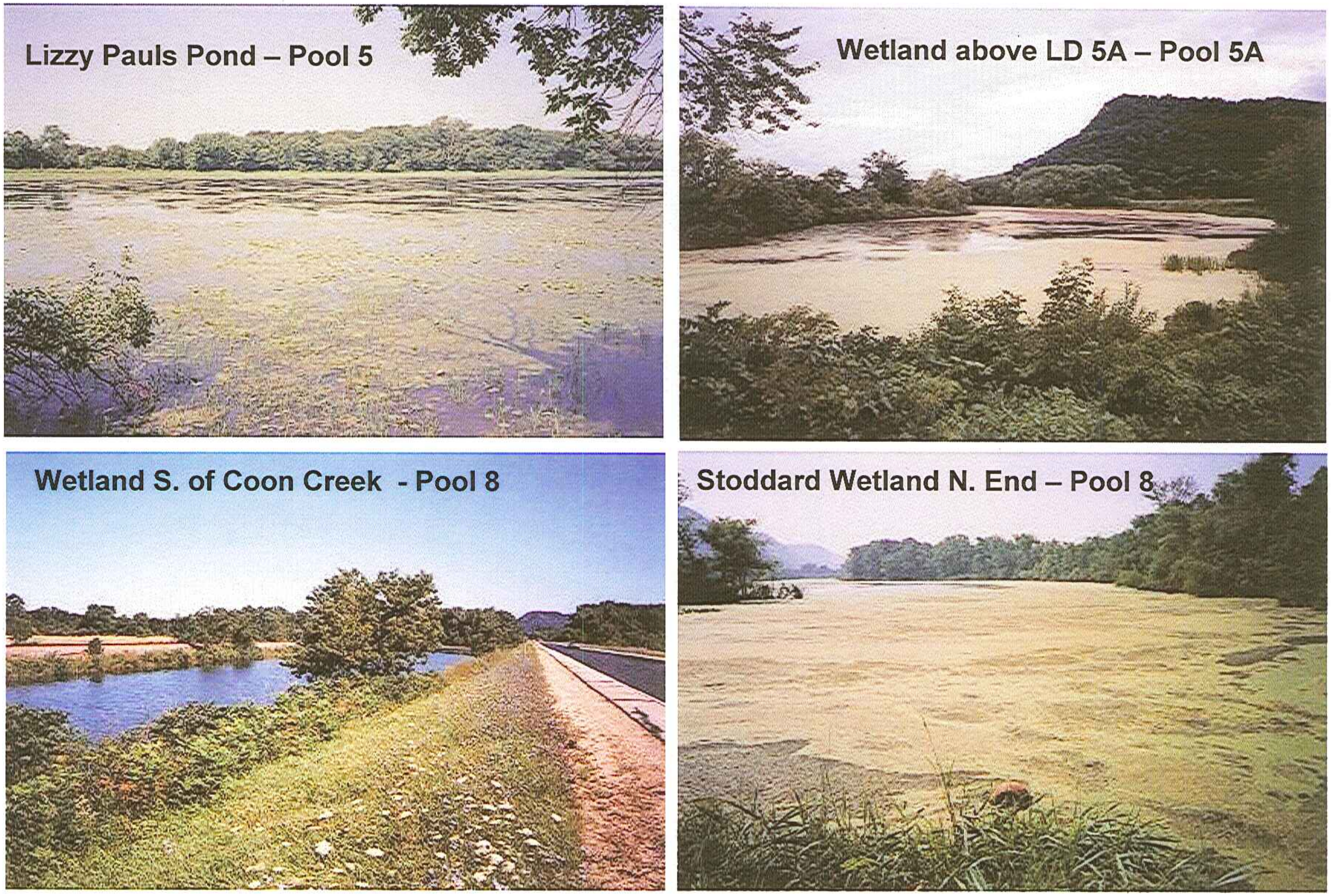


Figure 1. Continued – Detailed site maps.


Lizzy Pauls Pond – Pool 5

Wetland above LD 5A – Pool 5A

Wetland S. of Coon Creek - Pool 8

Stoddard Wetland N. End – Pool 8

Figure 2. Photographs of four study sites in the Mississippi River floodplain in navigation pools 5, 5A and 8.

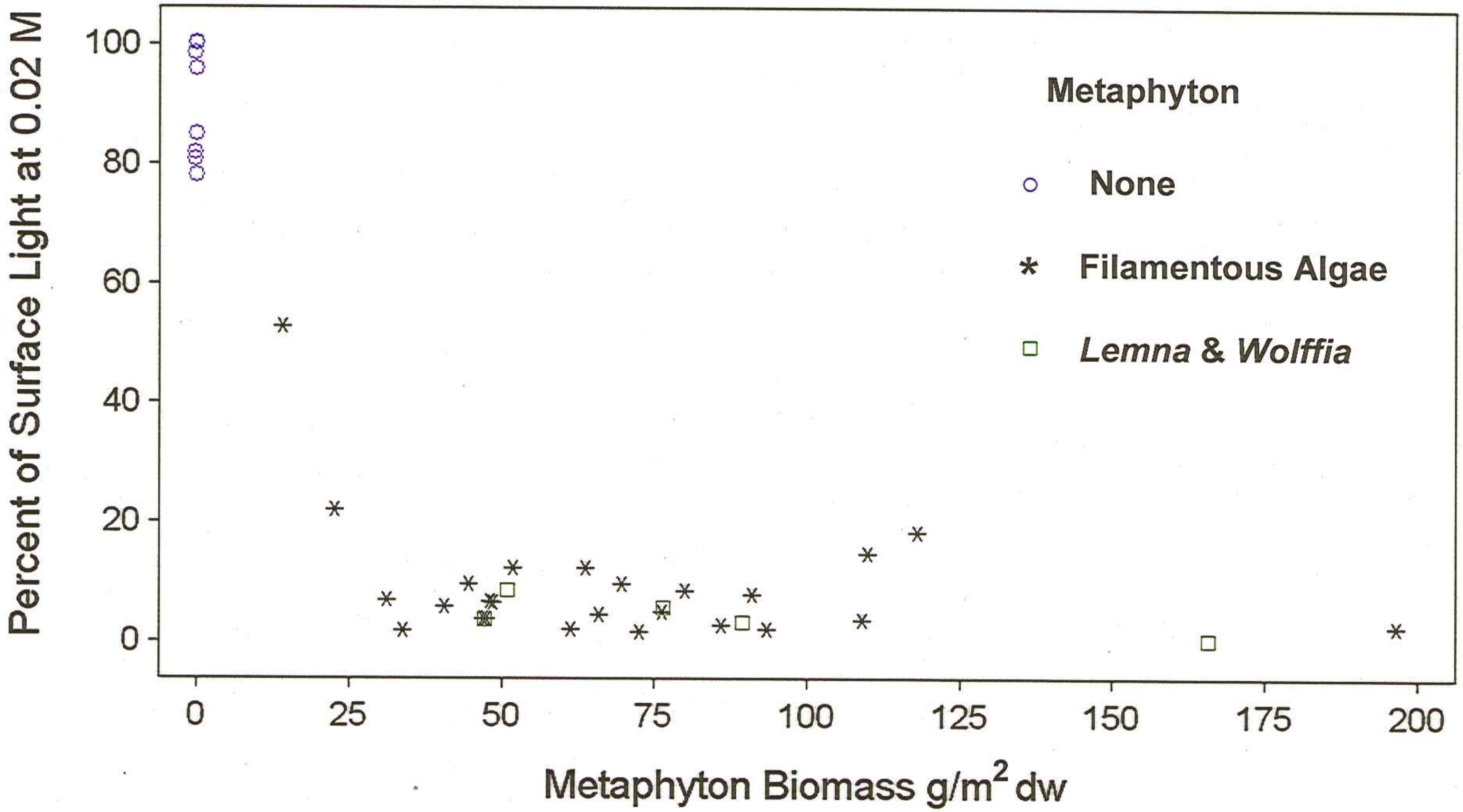


Figure 3. Underwater light measurements collected at 0.02 m depth in Mississippi River backwaters with varying metaphyton biomass. Light measurements were collected with a Li-Cor, Inc. surface quantum sensor (LI-190)and an underwater quantum sensor (LI-192).

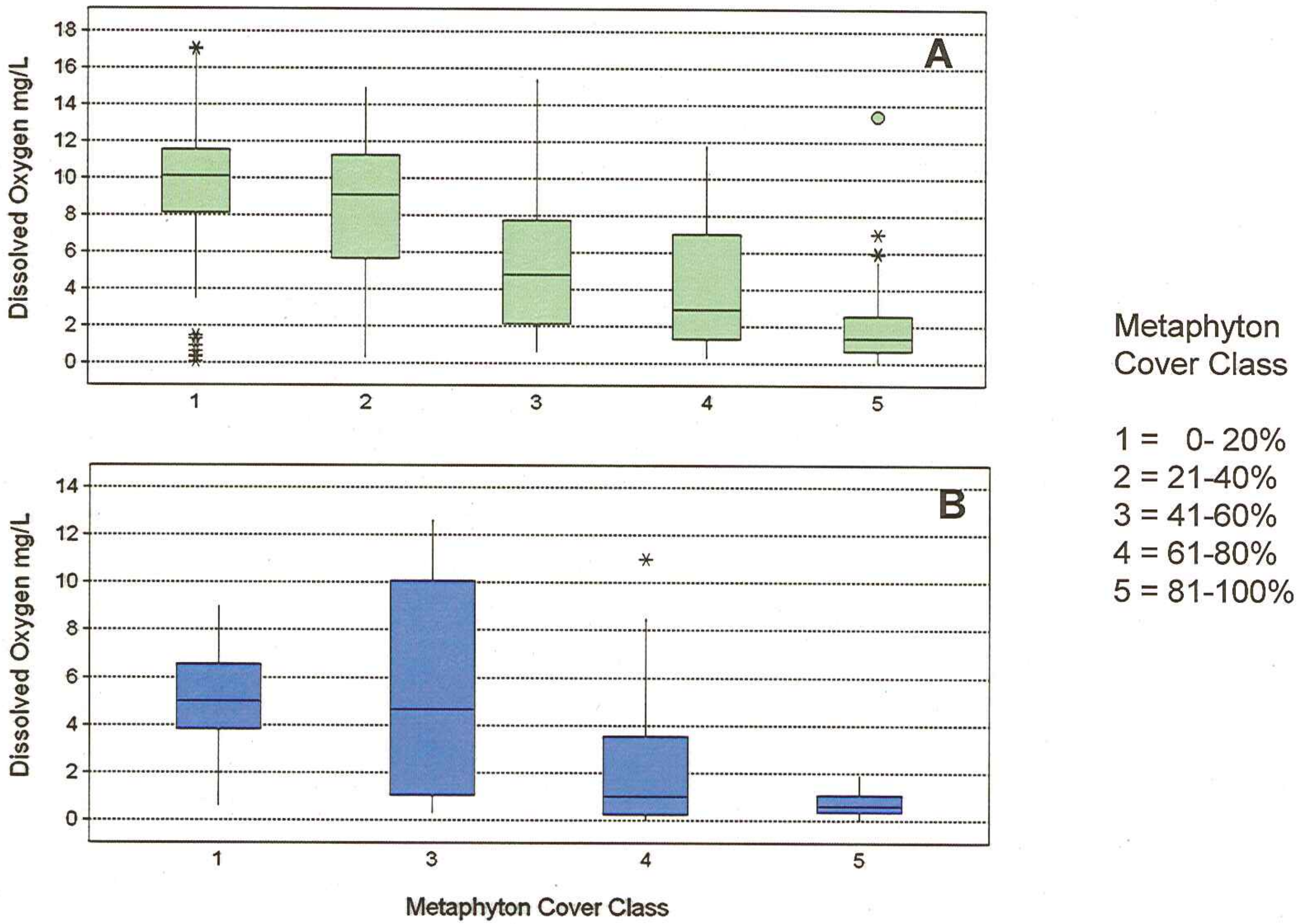


Figure 4. **A**. Dissolved oxygen levels versus *Lemna* cover measured during stratified random sampling of Pool 8 of the Mississippi River during July-August 2005-2007 by the Federal Long Term Resource Monitoring Program. **B**. Dissolved oxygen versus metaphyton cover (filamentous algae & duckweeds) made in this study.

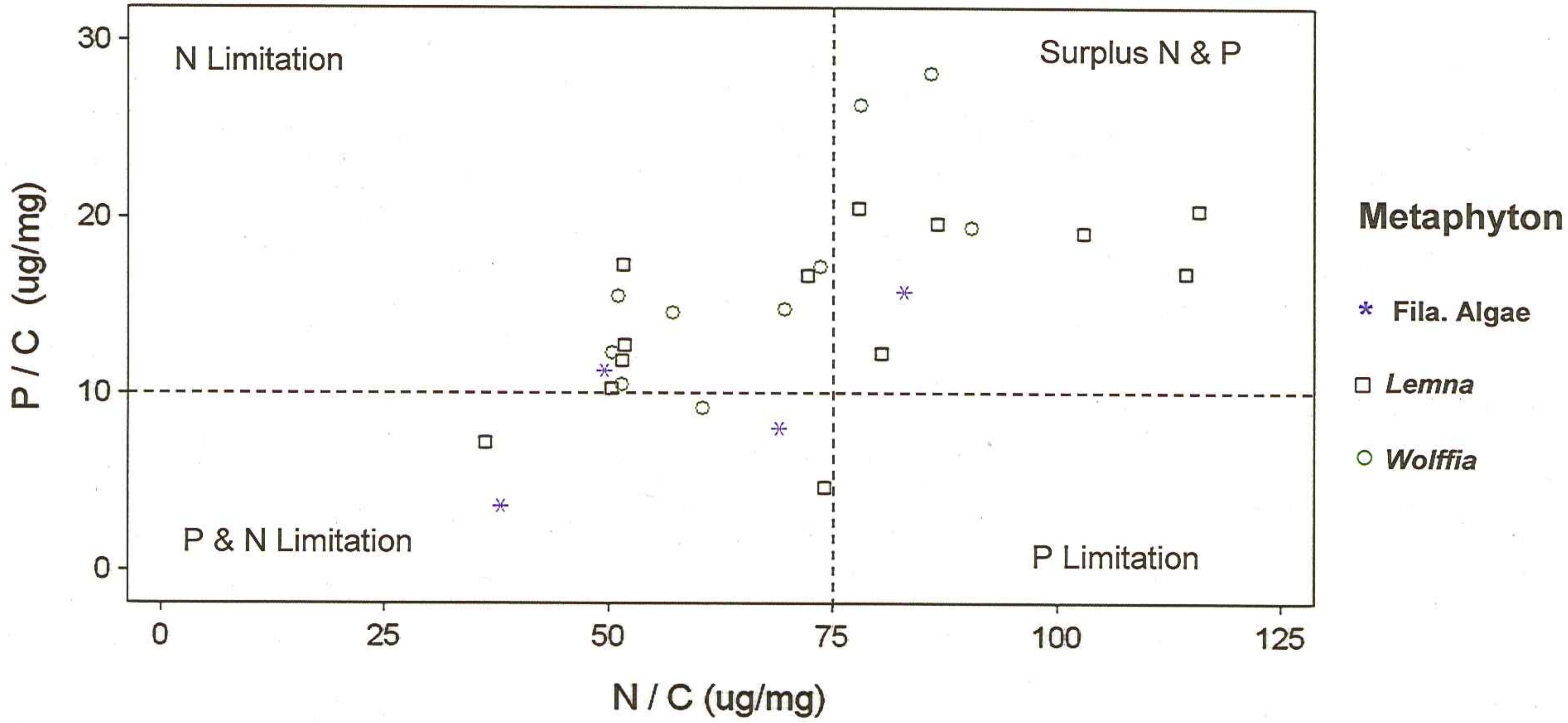


Figure 5. Phosphorus to carbon versus nitrogen to carbon composition ratios (mass basis) of metaphyton samples collected in Mississippi River backwaters bordering Wisconsin during July-August, 2005-2007. Thresholds for nitrogen limitation (N/C <75) and phosphorus limitation (P/C <10) were derived from Murkin et al. (1994).

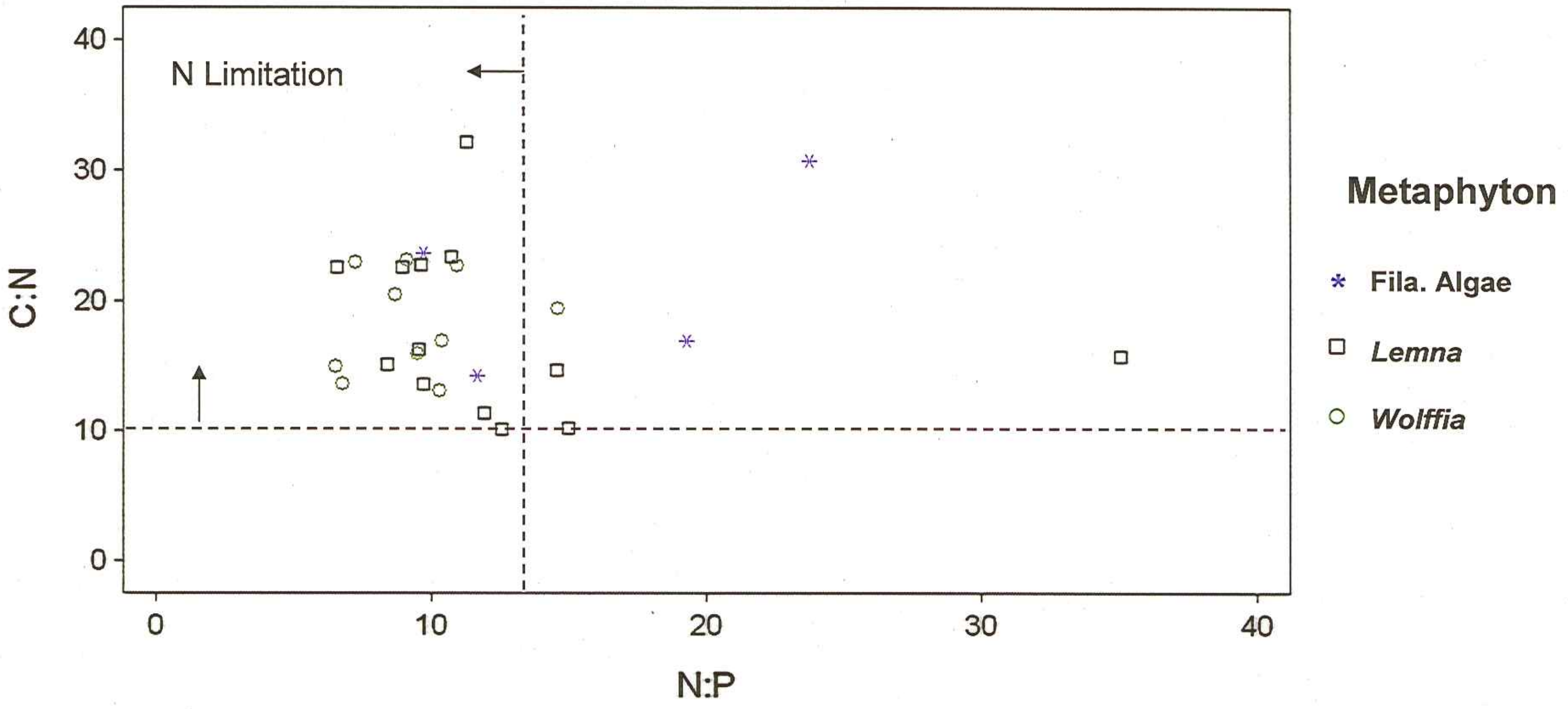


Figure 6. Carbon to nitrogen versus nitrogen to phosphorus composition ratios (atomic basis) of metaphyton samples collected in Mississippi River backwaters during July and August, 2005-2007. Threshold for N limitation (C:N >10 and N:P <13) derived from Hillebrand and Sommer (1999).

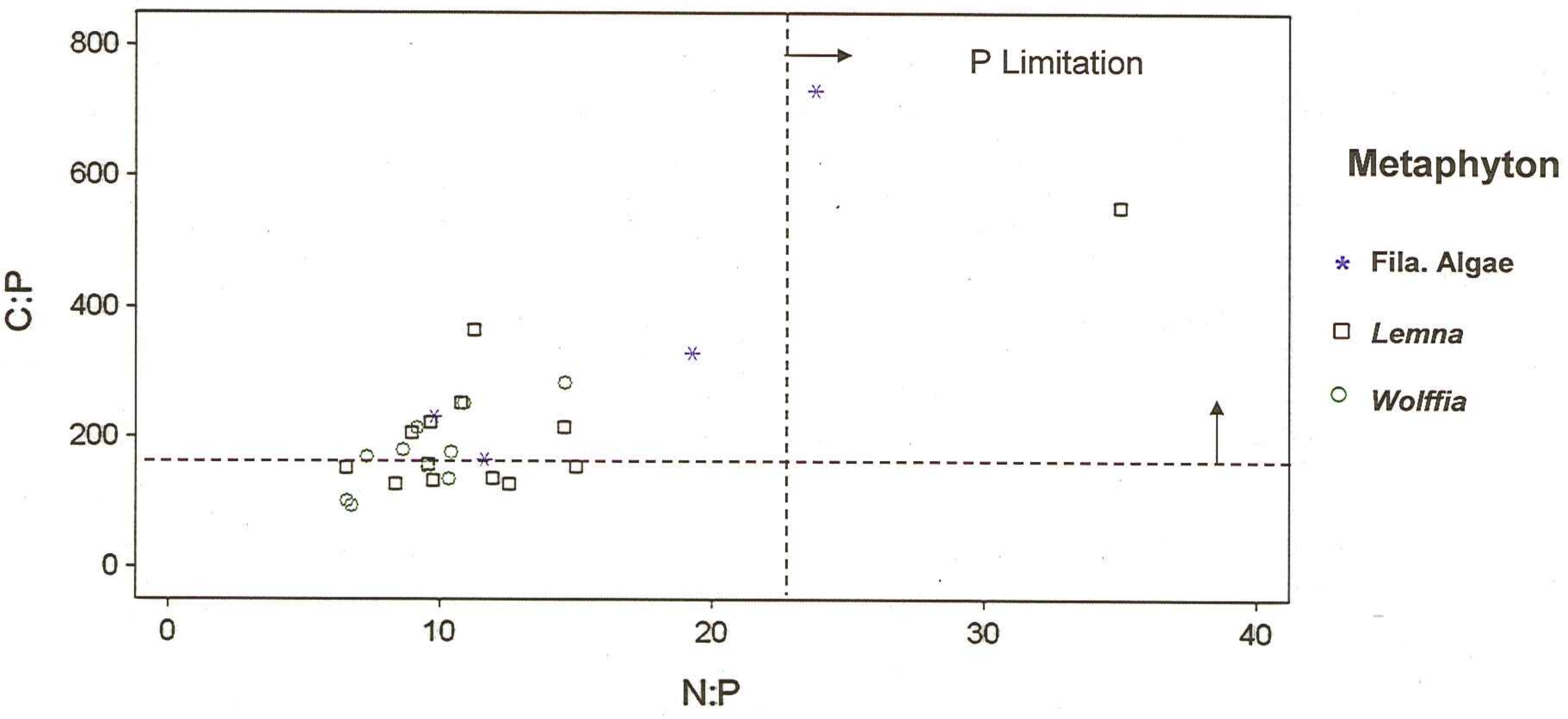


Figure 7. Phosphorus to carbon versus nitrogen to phosphorus composition ratios (atomic basis) of metaphyton samples collected in Mississippi River backwaters during July and August, 2005-2007. Threshold for P limitation (C:P > 180 and N:P > 22) derived from Hillebrand and Sommer (1999).

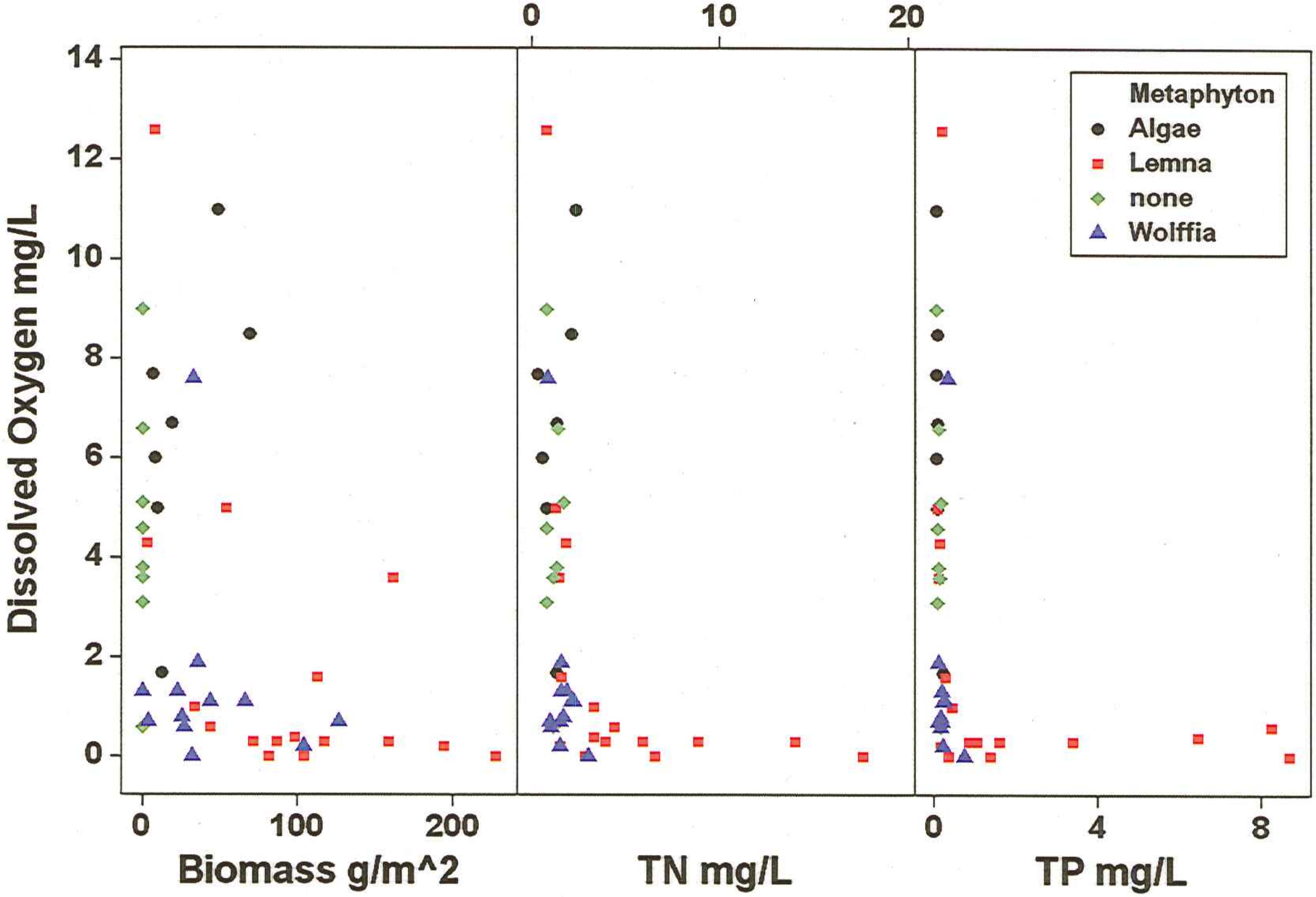


Figure 8. Mid-day dissolved oxygen versus metaphyton biomass (dry weight), total nitrogen and total phosphorus in Mississippi backwaters in Wisconsin from Pool 5 to Pool 9 during late July and early August 2003-2007.

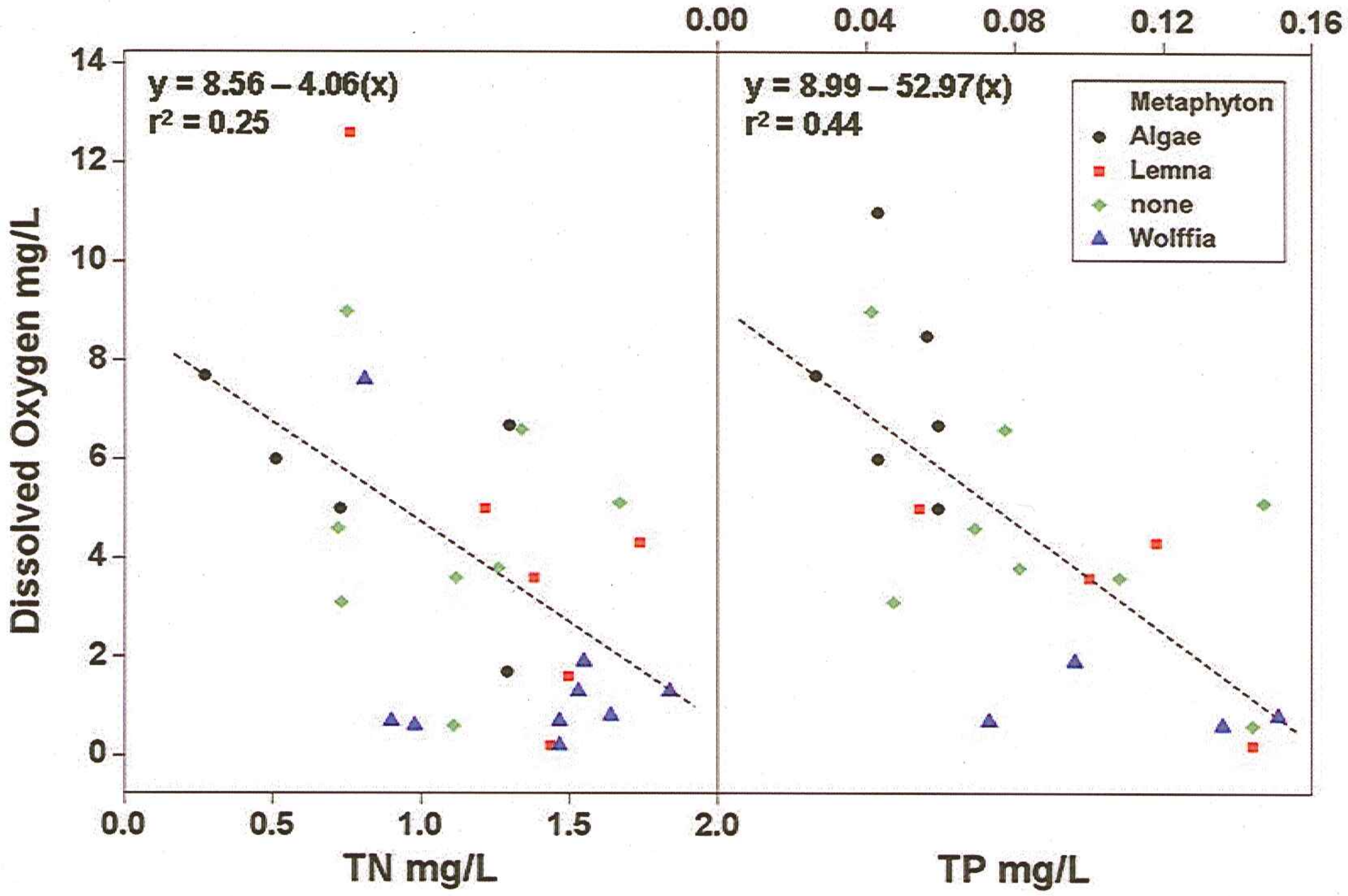


Figure 9. Linear regression analysis of dissolved oxygen versus total nitrogen and dissolved oxygen versus total phosphorus of Mississippi River backwaters along Wisconsin from Pools 5 to 9 during late July and early August 2003 to 2007. Note total nitrogen values > 2 mg/L and total phosphorus values > 0.16 mg/L were excluded from the analysis.

Table 1. Description of study sites located in Pools 5 to 9 of the Mississippi River.

Site Name	Site No.	Pool	Lat (deg.) NAD27	Lon (deg.) NAD27	N[1]	Period	Meta[2] Cover	Site Description
Lizzy Pauls Pond near wayside along HWY 35	063074	5	44.26235	-91.86326	5	2003-07	1	A shallow wetland isolated from the river with minimal impacts from pollution sources.
Wetland above Lock & Dam 5A along Hwy 35 - South End	063076	5A	44.09073	-91.66733	4	2003-06	4	A shallow wetland borded on the west by Burrlington Northern RR and HWY 35 on the East
Brice Prairie Channel 1/4 mi. above Lake Onalaska	323132	7	43.93578	-91.31517	3	2005-07	4	An abandoned distributary channel of Black River adjacent to Lake Onalaska. Substantial riparian residential development along NE bank
Wetland on west side of Pettibone Park - La Crosse, WI	323129	8	43.82048	-91.26766	4	2003-06	3	An isolated wetland withing floodplain forest adjacent to HWY 14/16
Wetland near Goose Island access road and HWY 35	633168	8	43.72833	-91.20166	4	2003-06	5	A small isolated shallow wetland at access road to Goose Island Park. Receives surface drainainge from development on east side of HWY 35.
Wetland S. of Goose Island Road near first road culverts	633170	8	43.72738	-91.20687	3	2003-06	5	A remote backwater adjacent to the Goose Island access road
Stoddard Islands backwater Stoddard, WI	10012517	8	43.67019	-91.22823	1	2006	4	A backwater area within the Stoddard Islands habitat project restoration area
Stoddard wetland north end Stoddard, WI	633166	8	43.65830	-91.22125	5	2003-07	5	A shallow isolated wetland near the village of Stoddard's wastewater treatment plant outfall. The outfall was relocated in the fall of 2005.
Stoddard wetland south end Stoddard, WI.	633167	8	43.64887	-91.22377	5	2003-07	4	A shallow isolated wetland south of the village of Stoddard, WI. Wetland had received municipal effluent prior to the fall of 2005.
Wetland S. of Coon Creek along HWY 35	633169	8	43.63917	-91.21283	4	2003-06	1	An isolated wetland bordering HWY 35 on east and the Burlington Northern RR on the West.
Desoto Pond along HWY 35 Desoto, WI	123075	9	43.41333	-91.19300	4	2003-06	1	An isolated backwater cutoff off from the river during construction of the Burlington Northern RR and HWY 35. The backwater is influeced by an artesian well.

[1] Number of Samples.

[2] Relative metaphyton cover: 1 = ≤20%, 2 = 21-40%, 3 = 41-60%, 4 = 61-80%, 5 = >80%.

Table 2. Summary of water chemistry and metaphyton biomass measurements in wetlands and backwaters in the Mississippi River along Wisconsin's floodplain from Pool 5 to 9. Samples were collected during late July and early August for years 2003 to 2007. A Kruskal-Wallis AOV was used to test for significant differences between sites. Values significant at $P<0.05$ are in bold.

Measurement	**Group 1 - Reference Sites[1]**					**Group 2 - All Other Sites[2]**					**Percent Difference in Medians**	**Kruskal-Wallis AOV P**
	N	**Median**	**Mean**	**Min.**	**Max.**	**N**	**Median**	**Mean**	**Min.**	**Max.**		
Depth m	13	0.6	0.7	0.5	0.9	30	0.6	0.7	0.3	1.8	0	0.218
Dissolved Oxygen mg/L	13	5.0	5.1	0.6	9.0	30	0.8	2.2	0.0	12.6	-85	**0.001**
Temperature C	13	26.4	25.5	22.0	29.3	30	23.6	23.9	21.0	26.6	-11	**0.018**
Sp. Conductivity uS/cm	13	471	474	295	715	30	426	554	245	1363	-10	0.958
pH	10	8.0	8.2	7.1	9.6	22	7.3	7.4	6.6	9.2	-9	**0.004**
Turbidity NTU	13	4.8	5.4	4.8	10.9	30	9.9	15.1	1.5	60.8	106	**0.046**
Total Suspended Solids mg/L	3	-	5.7	2.0	8.0	6	28.5	27.7	3.0	54		
Chlorophyll *a* ug/L	12	9.6	16.2	2.5	46.8	30	26.0	56.0	3.71	296	171	**0.013**
Ammonia-Nitrogen mg/L	13	0.015	0.02	< 0.013	0.05	30	0.027	0.05	< 0.013	4.95	80	**0.015**
Nitrite+Nitrate-Nitrogen mg/L	13	0.019	0.02	< 0.019	0.03	30	0.019	0.17	< 0.019	1.70	0	0.264
Inorganic Nitrogen mg/L	13	0.04	0.04	< 0.03	0.07	30	0.08	0.69	< 0.03	4.95	113	**0.002**
Total Kjeldahl-Nitrogen mg/L	13	1.08	1.00	0.25	1.72	30	1.57	3.22	0.63	17.6	45	**0.003**
Total Organic Nitrogen mg/L	13	1.07	0.98	0.24	1.70	30	1.55	2.70	0.60	12.7	45	**0.003**
Total Nitrogen mg/L	13	1.11	1.02	0.27	1.74	30	1.95	3.39	0.76	17.6	76	**0.000**
Dissolved Phosphorus[3] mg/L	13	0.007	0.011	< 0.002	0.028	30	0.029	0.540	< 0.002	7.02	307	**0.013**
Particulate Phosphorus mg/L	13	0.053	0.068	0.021	0.140	30	0.175	0.663	0.037	6.31	230	**0.000**
Total Phosphorus mg/L	13	0.069	0.078	0.026	0.147	30	0.204	1.20	0.043	8.68	195	**0.000**
Inorg. N / Dissolved P Ratio	13	4.9	8.1	2.1	23.5	30	4.7	37.0	0.2	573	-5	0.420
Total N / Total P Ratio	13	12.4	13.7	7.7	22.0	30	7.4	10.2	0.5	54.2	-40	**0.002**
Dissolved Silica mg/L	12	4.9	6.660	2.370	19.600	27	11.7	11.5	1.04	24.6	140	**0.015**
Metaphyton biomass g/m^2 dw	13	0.0	3.6	0.0	18.8	30	59.9	73.6	0.5	228	infinite	**0.000**

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[1] Sites where metaphyton cover was ≤ 20% (See Table 1).

[2] Sites where metaphyton cover was > 20% (See Table 1).

[3] Soluble reactive phosphorus.

Table 3. Summary of water chemistry measurements for different metaphyton types in wetlands and backwaters in the Mississippi River along Wisconsin's floodplain from Pool 5 to 9. Samples were collected during late July and early August for years 2003 to 2007. A Kruskal-Wallis AOV was used to test for significant differences between sites. Values significant at P<0.05 are in bold.

Measurement	Dominant Metaphyton Type								Kruska-Wallis AOV
	None		Algae		Lemna		Wolffia		
	N	Median	N	Median	N	Median	N	Median	P
Depth m	8	0.6	7	0.6	16	0.5	12	0.6	0.551
Dissolved Oxygen mg/L	8	4.2 **ab**	7	6.7 **a**	16	0.4 **c**	12	1.0 **bc**	**0.000**
Temperature C	8	26.2	7	24.1	16	23.9	12	23.9	0.489
Sp. Conductivity uS/cm	8	403	7	431	16	508	12	437	0.344
pH	7	7.9	5	8.0	11	7.2	9	7.4	**0.031**
Turbidity NTU	8	4.5	7	3.6	16	11.8	12	10	**0.045**
Total Suspended Solids mg/L	2	-	2	-	2	-	3	-	
Chlorophyll *a* ug/L	8	9.6	7	12.3	16	34.8	12	24.2	**0.032**
Ammonia-Nitrogen mg/L	8	0.018	7	0.015	16	0.302	12	0.019	**0.007**
Nitrite+Nitrate-Nitrogen mg/L	8	0.019	7	0.022	16	0.019	12	0.019	0.399
Inorganic Nitrogen mg/L	8	0.04 **b**	7	0.05 **ab**	16	0.53 **a**	12	0.04 **b**	**0.009**
Total Kjeldahl-Nitrogen mg/L	8	1.09 **bc**	7	0.63 **c**	16	3.00 **a**	12	1.52 **ab**	**0.000**
Total Organic Nitrogen mg/L	8	1.08 **bc**	7	0.61 **c**	16	2.79 **a**	12	1.50 **ab**	**0.000**
Total Nitrogen mg/L	8	1.11 **bc**	7	1.29 **b**	16	3.28 **a**	12	1.54 **ab**	**0.001**
Dissolved Phosphorus[1] mg/L	8	0.012 **b**	7	0.005 **b**	16	0.112 **a**	12	0.010 **ab**	**0.003**
Particulate Phosphorus mg/L	8	0.057 **bc**	7	0.046 **c**	16	0.435 **a**	12	0.170 **ab**	**0.001**
Total Phosphorus mg/L	8	0.079 **bc**	7	0.056 **c**	16	0.627 **a**	12	0.180 **ab**	**0.000**
Inorg. N / Dissolved P Ratio	8	4.2	7	17.0	16	2.8	12	4.7	0.341
Total N / Total P Ratio	8	13.5 **a**	7	12.4 **a**	16	5.8 **b**	12	8.6 **b**	**0.003**
Dissolved Silica mg/L	7	7.3	7	6.4	13	11.7	12	11.7	0.227
Metaphyton biomass g/m^2 dw	8	0 **b**	7	12.4 **ab**	16	92.4 **a**	12	32.6 **a**	**0.000**

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[1] Soluble reactive phosphorus.

Table 4. Spearman rank correlations of water chemistry and metaphyton measurements collected in wetlands and backwaters in the Mississippi River along Wisconsin's floodplain from Pool 5 to 9. Samples were collected during late July and early August for years 2005-2007. Values significant at $P<0.05$ are in bold.

Measurement	Metaphyton Biomass	Metaphyyton Cover	Chlorophyll *a*	Total N	Total P
Depth m	-0.210	-0.162	-0.382	-0.353	**-0.452**
Dissolved Oxygen mg/L	**-0.554**	**-0.587**	**-0.675**	**-0.612**	**-0.657**
Temperature C	-0.327	-0.234	0.086	-0.054	-0.109
Sp. Conductivity uS/cm	0.358	0.261	**0.424**	**0.600**	0.375
pH	**-0.572**	**-0.656**	**-0.609**	**-0.607**	**-0.433**
Turbidity NTU	**0.393**	**0.468**	**0.830**	**0.636**	**0.604**
Chlorophyll *a* ug/L	**0.387**	**0.489**	-	**0.683**	**0.577**
Ammonia-Nitrogen mg/L	**0.433**	0.254	0.270	**0.578**	**0.478**
Nitrite+Nitrate-Nitrogen mg/L	-0.144	0.158	0.263	0.214	-0.014
Inorganic Nitrogen mg/L	0.262	0.364	**0.581**	**0.632**	**0.454**
Total Kjeldahl-Nitrogen mg/L	**0.580**	**0.602**	**0.672**	**0.892**	**0.817**
Total Organic Nitrogen mg/L	**0.578**	**0.594**	**0.675**	**0.890**	**0.816**
Total Nitrogen mg/L	**0.679**	**0.642**	**0.683**	-	**0.729**
Dissolved Phosphorus[1] mg/L	0.246	**0.391**	0.234	**0.463**	**0.566**
Particulate Phosphorus mg/L	**0.650**	**0.558**	**0.669**	**0.761**	**0.951**
Total Phosphorus mg/L	**0.606**	**0.576**	**0.577**	**0.729**	-
Inorg. N / Dissolved P Ratio	-0.050	-0.225	0.073	-0.124	-0.306
Total N / Total P Ratio	**-0.432**	**-0.441**	**-0.442**	**-0.386**	**-0.871**
Dissolved Silica mg/L	0.297	**0.530**	**0.775**	**0.662**	**0.409**
Metaphyton biomass g/m^2 dw	-	**0.607**	**0.387**	**0.679**	**0.606**

[1] Soluble reactive phosphorus.

Table 5. Plant tissue analysis of metaphyton samples collected in wetlands and backwaters in the Mississippi River along Wisconsin's floodplain from Pool 5 to 9. Samples were collected during late July and early August for years 2005-2007. A Kruskal-Wallis AOV was used to test for significant differences between metaphyton types. Values significant at P<0.05 are in bold.

Plant Tissue Measurement	**Filamentous Algae**				***Lemna***				***Wolffia***				**Kruskal-Wallis AOV**
	N	**Mean**	**Min.**	**Max.**	**N**	**Mean**	**Min.**	**Max.**	**N**	**Mean**	**Min.**	**Max.**	**P**
N %	6	1.62 **b**	0.97	2.18	15	2.98 **a**	1.42	5.10	13	2.47 **ab**	1.75	3.80	**0.018**
P %	6	0.27 **b**	0.09	0.41	15	0.61 **a**	0.19	1.10	13	0.57 **a**	0.33	0.99	**0.008**
C %	4	29.1 **b**	25.4	33.9	13	39.8 **a**	36.0	44.5	10	36.4 **b**	33.5	39.1	**0.000**
Ca %	6	8.7 **a**	1.5	17.5	15	2.0 **ab**	0.9	3.8	13	1.8 **b**	1.1	3.1	**0.012**
K %	6	1.67	0.30	3.18	15	1.6	1.0	3.0	13	2.4	0.9	4.5	0.214
Na %	6	0.27 **b**	0.04	0.70	15	0.80 **ab**	0.35	1.60	13	1.22 **a**	0.69	2.09	**0.000**
Mg %	6	0.82	0.39	2.06	15	0.51	0.34	0.79	13	0.58	0.39	0.80	0.131
S %	4	0.54	0.17	1.09	10	0.45	0.22	0.58	10	0.38	0.26	0.65	0.347
C:N atomic	4	21.3	14.0	30.6	13	17.7	10.1	32.1	10	18.2	12.9	23.1	0.455
C:P atomic	4	362	164	728	13	213	126	551	10	173	91.9	281	0.173
N:P atomic	6	16.5	9.6	25.1	15	12.0	6.6	35.0	13	10.2	6.6	16.2	0.109
Al ug/g	6	1313	372	5392	15	461	6	1121	13	399	69	849	0.362
B ug/g	6	119 **b**	18.0	287	15	687 **a**	282	1568	13	475 **a**	127	1480	**0.000**
Cu ug/g	6	21.1	1.0	58.4	15	5.1	0.3	16.5	13	2.4	0.5	4.8	0.182
Fe ug/g	6	2913	1379	5150	15	2137	254	4184	13	2850	498	8332	0.434
Mn ug/g	6	2480 **ab**	438	4506	15	1416 **b**	854	1823	13	4064 **a**	1025	13144	**0.027**
Zn ug/g	6	52.1	7.9	101.0	15	28.3	12.1	39.6	13	23.3	10	29.0	0.365

Table 6. Proposed nutrient criteria for total phosphorus and total nitrogen for Mississippi River backwaters and wetlands.

	Nutrient Criterion		
Basis of Nutrient Criteria	**Total Phosphorus mg/L**	**Total Nitrogen mg/L**	**Comment**
Dissolved Oxygen			
5 mg/L	0.075	0.88	DO x TP Regression for TP < 0.16 mg/L DO = 9.012 - 53.34(TP) $r^2 = 0.484$
3 mg/L	0.113	1.37	
1 mg/L	0.150	1.86	DO x TN Regression for TN < 2 mg/L: DO = 8.564 - 4.056(TN) $r^2 = 0.250$
Metaphyton Cover			
≤ 20%	0.078 (0.069)	1.02 (1.11)	Average and (median)
≤ 60%	0.160 (0.081)	1.18 (1.11)	Average and (median)
Average	0.119 (0.075)	1.10 (1.11)	Estimate for ≤ 40% cover
Metaphyton Biomass			
0 g/m^2 dw	0.089 (0.079)	1.09 (1.11)	Average and (median)
< 10 g/m^2 dw	0.092 (0.077)	1.01 (0.90)	Average and (median)
< 25 g/m^2 dw	0.101 (0.079)	1.09 (1.11)	Average and (median). Substantial light attenuation occurs at biomass exceeding this value.