Nitrate-N trends in Mississippi and Atchafalaya River Basin Watersheds: Exploring correlations of watershed features with nutrient transport components 2000-2020.

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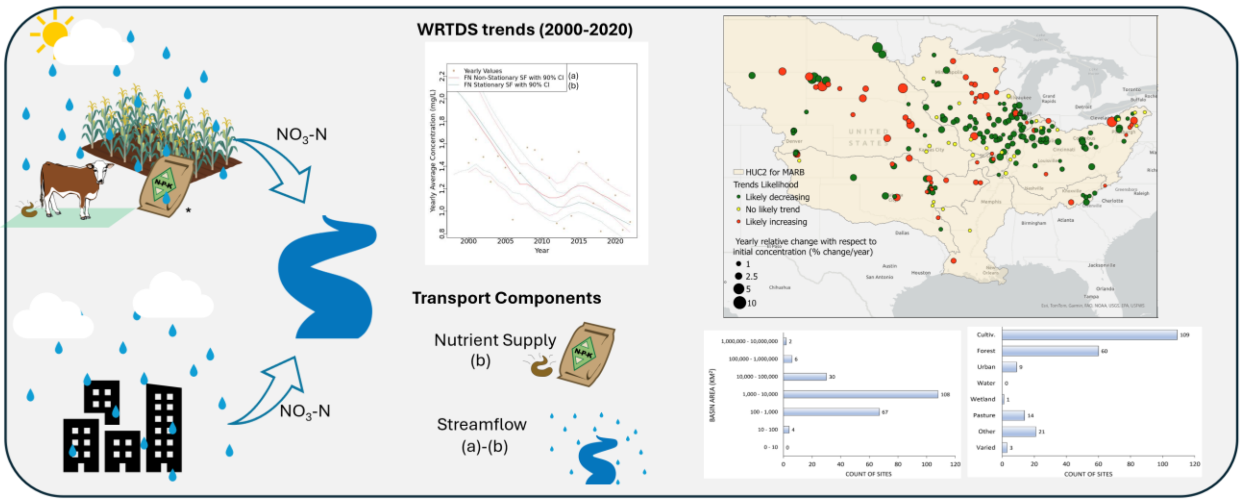
1. Abstract

Nutrient reduction strategies in the Mississippi and Atchafalaya River Basin (MARB) have been implemented since mid-2010’s to attenuate the impacts of non-point source pollution to the Gulf of Mexico. Of all nutrients, nitrate represents the largest threat due to its extended presence throughout the basin and its high solubility in water. To evaluate the performance of state reduction strategies, long-term changes of riverine nitrate should be identified. The objective of this study was to estimate the flow-normalized (FN) nitrate-N concentration and yield trends for the 2000-2020 period across the MARB. A harmonization and in-depth screening of paired nitrate-N and streamflow datasets resulted in a robust water quality monitoring network of 217 sites. Trends magnitude and likelihood were computed using the Weighted Regression on Time, Discharge, and Season (WRTDS) coupled to a bootstrap test, and trends results were correlated with basin features and initial values. Nutrient supply and flow components of trends were computed through the stationary and non-stationary flow normalization. Results indicated that 59.4% of the 217 sites had likely decreasing trends, while 27.6% likely increased, and the remaining 12.9% had no likely change detected. Reductions in riverine FN nitrate-N were clustered in the north central region of the MARB, with watersheds dominated by cultivated cropland having relatively high FN concentrations and yields in 2000 followed by likely downward trends. For the vast majority of sites, the streamflow component contributed to increased FN nutrients, but the nutrient supply component was more dominant than the streamflow component.

Keywords: Nutrient trends, Nitrate-N, Streamflow, Nutrient supply, Mississippi and Atchafalaya River Basin, Weighted Regressions on Time, Discharge, and Season model (WRTDS).

2. Graphical Abstract



*Maize, Fertilizer, Cow, Feces, & Weir illustrations from Jane Thomas, Tracey Saxby & Jane Hawke at Integration and Application Network (ian.umces.edu/media-library)

3. Introduction

Anthropogenic nutrient pollution from cultivated and urban land in the Mississippi and Atchafalaya River Basin (MARB) is the main cause of the 16,000 km^2 hypoxic zone in the northern Gulf of Mexico (Piske and Peterson, 2020). Of all nutrients transported to the Gulf of Mexico, nitrate is the principal cause of the low-oxygen levels at the Gulf (Rabalais and Turner, 2019). Sources of this nutrient include inorganic N fertilizers, manure, soil organic matter, biological N fixation, municipal wastewater and atmospheric deposition (McIsaac et al., 2002, Stackpoole et al., 2021). The MARB has one the most productive farming regions worldwide (Goolsby and Battaglin, 2001), making cultivated land the most significant source of nitrate. The implementation of tile drainage in cultivated land has also played a main role on nitrate transport within the MARB (David et al., 2010).

Federal, state, and local agencies have implemented nutrient reduction strategies to reduce the amount of pollutants entering streams (Mississippi River Gulf of Mexico Watershed Nutrient Task Force, 2008). Analyses of long-term changes in the nitrate loads and concentrations in streams across the MARB allow the identification of regions having the largest reductions and increases of nutrient contribution over time. Goolsby and Battaglin (2001) investigated the long-term change on the average nitrate flux in the MARB, finding 1980-99 nitrate-N flux to be three times larger than that of 1955-70. This increase coincided with increased use of N fertilizer as well as an increase in precipitation and river flow. Donner et al. (2002) used models to estimate that 25% of the increase in nitrate load was due to increased streamflow. Distinguishing the influences of changing nutrient source management from climate factors on river loads is an ongoing challenge.

To increase the amount of information extracted from riverine concentration time series, and reduce the impact of year-to-year streamflow variations on long-term trends analysis, the Weighted Regression on time, discharge, and season (WRTDS) was proposed (Hirsch et al., 2010). This method was applied to estimate nutrient trends up to 2012 for the MARB (Oelsner et al., 2017, Crawford et al., 2019), and up to 2020 for 110 USGS water quality monitoring sites across the U.S. (US Geological Survey, 2024). A further development of the WRTDS method allowed the computation of trends considering the non-stationarity of flow (Choquette et al., 2019, Hirsch, 2018). This advancement permitted the separation of streamflow driven transport component from all other non-streamflow components that could impact the fate and transport of nutrients (Murphy and Sprague, 2019), hereafter named nutrient supply transport component.

Transport and retention of nutrients are mainly controlled by the advective processes occurring as water flows throughout the watershed, and the availability of these non-point source pollutants in watershed compartments (Speir et al., 2021). Both flow and nutrient supply components are influenced by watershed features. Land use, soil, topography, and climatological conditions determine rainfall to runoff conversion, infiltration, and evapotranspiration processes, all of these influencing water volumes and residence times (Goyette et al., 2019). Land use is the main driver of nutrient supply, linked to fertilizer and manure application and legacy nitrogen from current and historical agricultural land (Stackpoole et al., 2021), and to point sources from urban areas. Furthermore, basin area influences the complexity of the river system being studied. Small catchments often allow the clear identification of sources contributing to measured loads, as opposed to large watersheds (i.e., larger than a couple of hundreds of square kilometers), in which diverse interconnected systems regulate the fate and delivery of nutrients at the outlet (Alexander et al., 2002), making nutrient reduction strategies tougher to implement and assess.

The objective of this paper is to conduct a methodologically consistent trend analysis of riverine Nitrate-N concentration and yield (load/drainage area) across the MARB from 2000 to 2020 and estimate the relative influences of streamflow and nutrient supply component on trends results using screened nitrate-N and streamflow datasets across the MARB. The correlation of trends results and watershed features (i.e., drainage area, relief, upstream dam storage, and historical dominant land use) was analyzed. An analogous analysis was performed for Total Phosphorus (Botero-Acosta et al., In preparation).

4. Materials and Methods

The trend period selected for this analysis provided the largest number of sites meeting data requirements to allow for a meaningful study across the MARB. A 20-year trends period (2000-2020) with 2-year additional periods at starting and ending periods, to improve the accuracy for 2000 and 2020 estimations, was used. In addition, formal state nutrient loss reduction strategies were implemented from mid-2010’s, making this period of particular interest to evaluate the performance of the selected approaches.

4.1. Data harmonization and screening

A riverine nitrate-N concentration dataset was compiled from all records having nitrate or nitrate+nitrite concentrations in filtered or unfiltered water samples in the US Water Quality Portal (National Water Quality Monitoring Council et al., 2021). This definition was based on the reported minor concentrations of nitrite with respect to nitrate and the marginal difference between filtered and unfiltered samples (Oelsner et al., 2017). Records from sites linked to the same stream segment were unified to increase the time series length at these locations. This process was done through the COMID feature of the National Hydrograph Dataset Plus (EPA, 2022). Nitrate-N data was harmonized and screened. The harmonization of the observed dataset plays a major role when analyzing historical changes of nutrient loads (Sprague et al., 2017). Representatives and documentation from the various reporting agencies were consulted to clarify methods and units of ambiguous records. For nitrate data, 12% of records with ambiguous form (elemental or molecular) or units were clarified by contacting the reporting organization, while 32% were solved through the reported analytical method.

Harmonized concentration data were included in trend calculations only after various screening criteria were met. Initially, a record screening identified and solved duplicates and censored records. Missing, zero, negative, and outlier records were removed from the analysis, as well as composite, control and field analyzed samples. Subsequently, site screening removed sites that had more than 50% of left-censored data, less than 70% quarterly coverage for the 2000-20 period, and less than 10% of decadal WQ data on days with high flow regime (SF>85 percentile).

Streamflow (SF) sites located upstream or downstream of WQ sites were selected from the USGS gage network when basin areas had a maximum difference of 10% and no dams were located in between the sites. Data of dams built before 2013 were extracted from EPA (2022).

SF data screening identified missing, zero and negative flow values. Missing records were filled for years having no more than 3 consecutive and 30 total missing records using the FillMiss function (USGS, 2016). All SF sites with no consecutive records for the 1998-2022 water year periods were discarded. From the screening and harmonization procedure, 217 sites were identified for which flow normalized nitrate concentration and load trend analyses between 2000 and 2020 were conducted.

4.2. Nitrate-N trends

The Weighted Regression on Time, Discharge and Season (WRTDS) (Hirsch et al., 2010) method was implemented to compute the flow normalized (FN) concentration and loads from observed data. The performance of the WRTDS method was evaluated through the Pearson correlation coefficient, the extrapolation metric and the flux bias statistics. In addition, a visual inspection of residuals was performed to remove sites with step changes in concentrations, not reproduced by the WRTDS method (Oelsner et al., 2017).

Flow normalization was calculated by two methods: 1) normalized to average flow distribution observed for the period of observation (stationarity assumption); and 2) normalized to a moving window of flow distributions (non-stationarity assumption or generalized normalization). The difference between these two estimates is considered the change due to non-stationary flow. The rest of the change is due to all other factors, including changes in point and non-point sources, legacy nutrients, land cover and management. For brevity, Murphy and Sprague (2019) use the term “management component”, which may not adequately convey the role of all other factors such as legacy nutrients and inaccuracies in WRTDS flow normalized estimates, and may understate the impacts of management practices on water flow. Rather than “management component” we use the term “nutrient supply component”, but acknowledge that we have not quantified specific nutrient supplies such as fertilizer inputs or point sources, etc.

Our use of the term “nutrient supply” is a catchall term that refers to all factors other than changes in average streamflow, which is a proxy for nutrient sources and sinks. All Nitrate-N concentration and yield values referred to in the results section correspond to flow normalized values.

Trends were determined by the difference between estimated FN concentration or load for 2020 and 2000. To reduce the impacts of streamflow random variations on resulting nutrient time series, the flow-normalized values of the obtained time series were used to estimate long term trends (Murphy and Sprague, 2019, Hirsch et al., 2010). WRTDS has been widely used to characterize trends of nutrient loads (e.g., McIsaac et al. (2023), US Geological Survey (2024)), and to estimate the impacts of management practices and flow on resulting trends (Murphy and Sprague, 2019). Analyses were conducted using the R statistical software program (R Core Team, 2017) and the EGRET R-package (Hirsch, 2018, Hirsch and De Cicco, 2015).

To compare trends results among sites with varied characteristics, concentration trends were weighted by the flow normalized concentration at the beginning of the trends period (2000) and divided it by the number of years, which provided a percent change of concentration per year (Murphy and Sprague, 2019). Likewise, load trends were weighted by the drainage basin area (USGS, 2024) to identify the yearly nutrient yield expressed in kilograms per square kilometer and year. Analyzing both relative and absolute changes will provide insights about basin specific processes impacting nutrient concentrations during the trends period as well as allow the identification of those basins that might be larger contributors of nutrient loads.

A likelihood-based approach, the block bootstrap method (Hirsch et al., 2015), was used to estimate the likelihood of the total trends for concentration and yield (values normalized under the non-stationary flow assumption). This method runs multiple replicates of WRTDS using randomly selected subsamples from the observed data and computed the fraction of replicates with positive trend (σ). A positive replicate fraction of 0.66 or larger would indicate a likely upward trend, while an estimate of positive replicates of less than 0.33 would indicate a likely downward trend, and fractions between 0.33 and 0.66 would correspond to no likely trend since similar number of replicates were found to have increasing and decreasing trend (Yates et al., 2022, Hirsch et al., 2015). Positive replicate fractions > 0.90 and <0.10 were considered highly likely upward and downward trends, respectively.

The dominance of one trend component vs the other (e.g., flow vs. nutrient supply) was established based on the factor of exceedance. A transport component was identified as dominant if it was at least 1.5 times the other component (using absolute values). The percentage of influence of each transport components (flow or nutrient supply) was computed as:

$$\%\ Influence\ component\ A = \frac{abs(\%\ change\ component\ A)}{abs(\%\ change\ component\ A) + abs(\%\ change\ component\ B)} \qquad \text{Eq.1}$$

Influence of Nutrient Supply and Streamflow components on load trends are equivalent to the influences of both components on yield trends.

4.3. Watershed features

Data for basins associated with the nitrate-N trends sites was analyzed along with the trends results. Basin area, land use, elevation, and upstream dam storage information were extracted from publicly available data. Dominant land uses for years 2001, 2011, and 2019 were identified from National Land Cover Dataset (NLCD) maps and the combined land use categories (Table 1) covering the largest area of the drainage basin (Stets et al., 2020). The dominant land use for the 2000-2020 trend period was identified for those basins having the same dominant land use category prevailing for the 2001, 2011, and 2019 NLCD maps. When the dominant category changed during the 2000-2020 period, the label “Varied” was assigned. In addition, changes of percentage coverage from 2001 to 2019 for each land use category were computed and correlated with riverine nitrate trends. A proxy of basin slope was included in our analysis as the relief per area unit (m/km^2). Relief was computed as the difference between the maximum and the minimum elevation within the basins draining to trends sites from a 5x5 degree DEM. Finally, dam storage information was extracted from the National Hydrography Dataset Plus (NHD-Plus) (EPA, 2022).

Table 1. National Land Cover Database (NLCD) categories grouped for watershed feature analysis.

Adapted land use categories	NLCD Category ID#	Description
Cultivated (tilled land)	82	Cultivated Crops -areas used to produce annual crops, such as corn, soybeans, vegetables, tobacco, and cotton, and perennial woody crops such as orchards and vineyards. Crop vegetation accounts for greater than 20% of total vegetation. This class also includes all land being actively tilled.
Forest	41	Deciduous Forest
	42	Evergreen Forest
	43	Mixed Forest
Urban	22	Developed, Low Intensity
	23	Developed, Medium Intensity
	24	Developed High Intensity
Water	11, 12	Water
Wetlands	90, 95	Wetlands
Pasture/Hay	81	Pasture/Hay
Other	21	Developed, Open Space- areas with a mixture of some constructed materials, but mostly vegetation in the form of lawn grasses.
	31	Barren Land (Rock/Sand/Clay)
	52	Shrub/Scrub
	71	Herbaceous

Watershed features were analyzed through Pearson correlation (r) matrices to evaluate the direction and the degree of collinearity between variables. The statistical significance of correlations was evaluated by testing the null hypothesis of no correlation with an alpha value of 0.05.

5. Results and discussion

5.1. Watershed Features

The basins associated with the 217 nitrate-N trends sites (Figure A- 1) were characterized as described in Section 4.3. Cultivated cropland was the dominant land cover for 50% of these sites, followed by forest (27.6%) (Figure 1, Table 2). It was found that 50% of sites had a Relief per area between 0 and 0.1 m km^{-2}, and a somewhat different 50% of sites had drainage areas between 1,000 and 10,000 km^{2}. Subdivisions of categories of relief per area, land cover and drainage areas for the sites are illustrated in Figure 1.

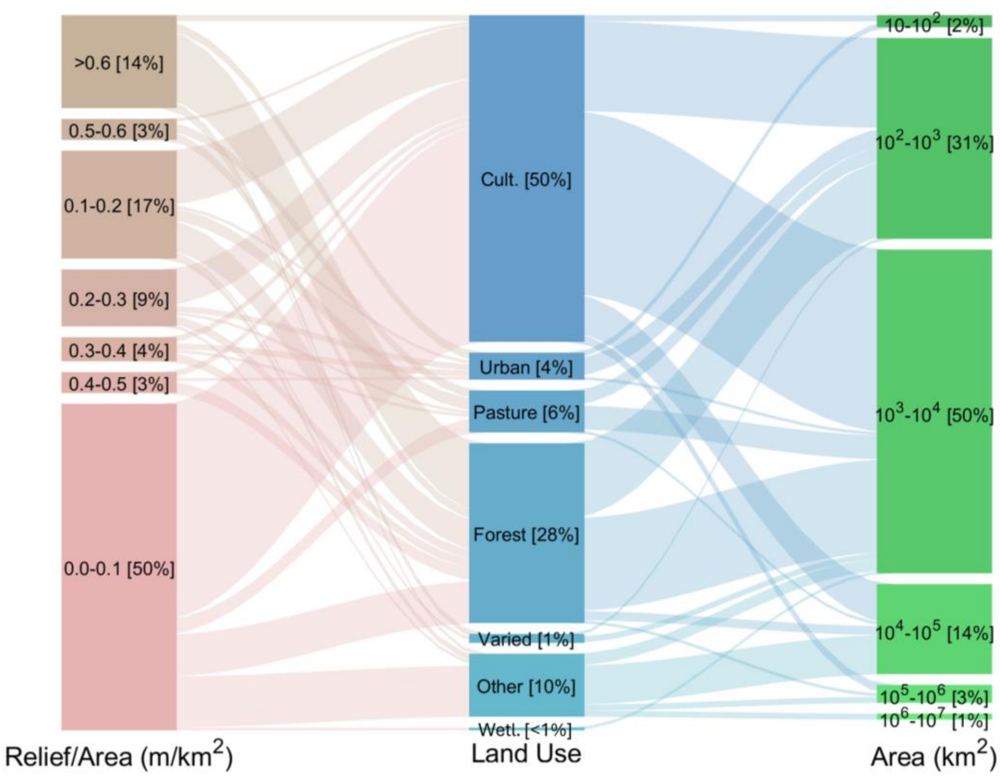


Figure 1. Main watershed features for the 217 nitrate-N trends sites and percentage of sites for each category.

5.2. Overall Trends

Flow normalized (FN) nitrate-N concentrations likely decreased at 59.4% of the 217 sites and increased at 27.6%, with no likely change detected for the remaining 12.9% (Table 2). FN yields likely decreased at 47% of sites, increased at 39.2% of sites and no trend was detected at 13.8% of sites. Reductions in FN nitrate-N concentration and yield were clustered in the north central region of the basin (Figure 2 and Figure 3), much of which is intensively cultivated cropland.

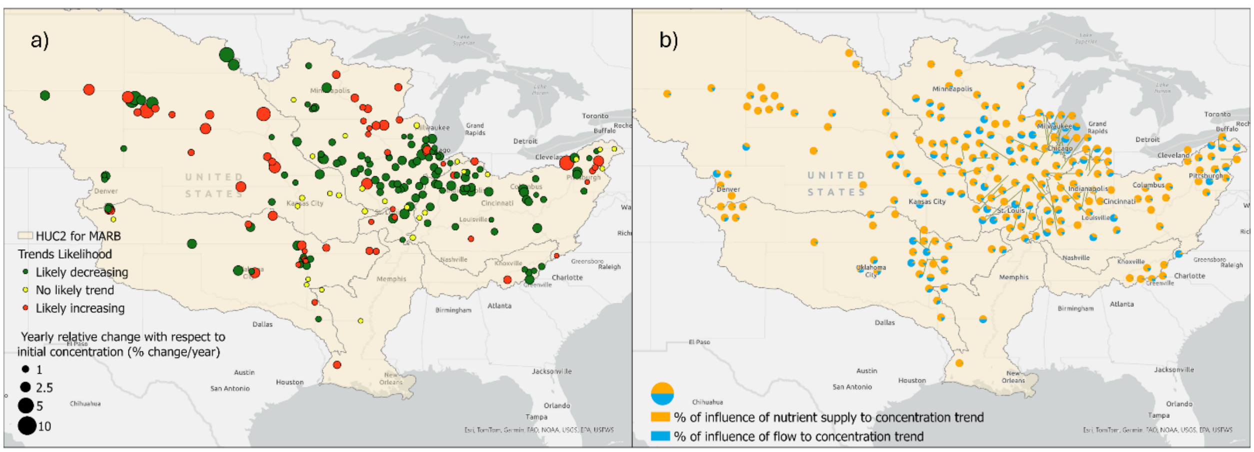


Figure 2. a) Nitrate-N concentration trends for the 2000-20 period (n=217). Magnitudes were set based on absolute values of total trends; colors were set based on trends likelihood. b) Percent influence of nutrient supply and flow components.

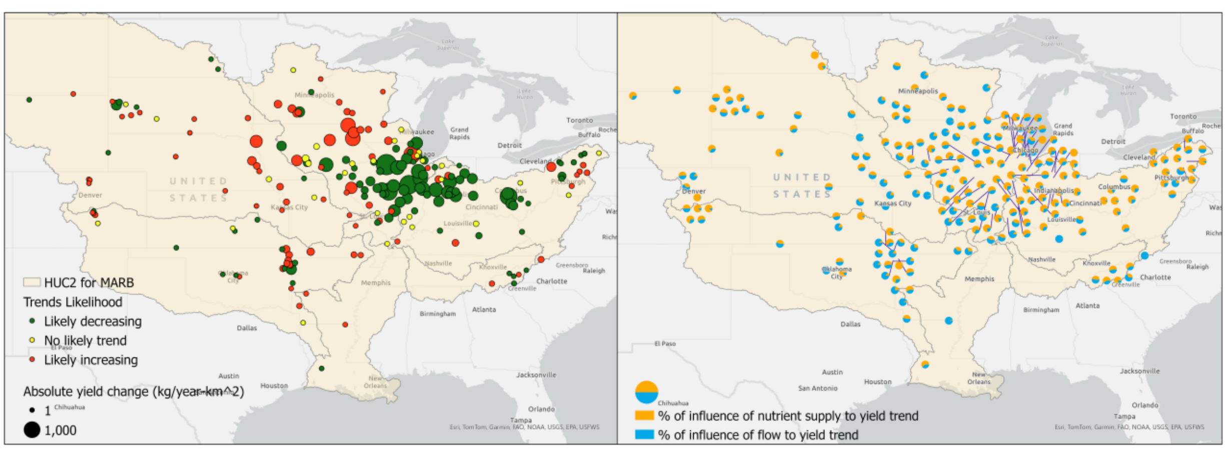


Figure 3. a) Nitrate-N yield trends for the 2000-20 period (n=217). Magnitudes were set based on absolute values of total trends; colors were set based on trends likelihood. b) Percent influence of nutrient supply and flow components.

Land cover for slightly more than half of the monitoring locations wase dominated by cultivated cropland (Figure 1). Of these sites, FN nitrate concentrations likely decreased in 73.4% of the sites and FN nitrate yield likely declined in 58.7%. Nationwide trends for the 2002-2012 period (Stets et al., 2020) reported decreasing nutrient concentration at 40% of the studied sites dominated by cropland. Our results shows an improvement possibly due to increased N use efficiency in corn production (Mueller et al., 2019, Haegele et al., 2013) and the implementation of nutrient reduction strategies since mid-2010's (Christianson et al., 2018). In contrast, for the watersheds dominated by forest cover FN concentration trends were more equally divided between likely increasing (43%) and decreasing trends (40%); and a greater percentage had likely increasing FN yield trends (56.7%) than decreasing (38.3%) (Table 2).

Sites with urban dominated basins (9 sites) showed a predominance on likely decreasing concentrations (77.8%) and yield (44.4%). This percentage of sites seems to be larger than the one reported by Stets et al. (2020) for urban basins in their national trend analysis for the 2002-2012 period (>60%), an indication of the success of implemented management efforts of the 1972 Clean Water Act for regulating point discharges that, as we evidenced, have continued to the 2000-2020 period at the MARB.

Table 2. Distribution of sites by trend likelihood and dominant land cover.

			% of sites for concentration trend categories			*% of sites for yield trend categories*		
Dominant Land use	**% of sites**	**Total area (km2)**	**Likely Increasing**	**Likely Decreasing**	**No likely Trend**	**Likely Increasing**	**Likely Decreasing**	**No likely Trend**
Varied (3)	1.4%	4932.7	33.3%	33.3%	33.3%	100.0%	0.0%	0.0%
Other (21)	9.7%	3916307.8	42.9%	47.6%	9.5%	38.1%	38.1%	23.8%
Pasture (14)	6.5%	43315.2	35.7%	50.0%	14.3%	64.3%	21.4%	14.3%
Urban (9)	4.1%	3380.7	22.2%	77.8%	0.0%	22.2%	44.4%	33.3%
Forest (60)	27.6%	426003.9	43.3%	40.0%	16.7%	56.7%	38.3%	5.0%
Cultiv. (109)	50.2%	1092838.1	14.7%	73.4%	11.9%	25.7%	58.7%	15.6%
Wetland (1)	0.5%	2563.7	100.0%	0.0%	0.0%	100.0%	0.0%	0.0%

MARB (217)	**100%**	**5489342 ***	**27.7%**	**59.4%**	**12.9%**	**39.2%**	**46.95%**	**13.8%**

Table note: **Total drainage area is larger than the MARB because there are overlapping drainage areas in basins with multiple monitoring locations. For instance, the drainage area of the Mississippi River at Thebes includes all the upstream basins, including the Missouri River at Herman*

Mean rates of change in FN nitrate-N concentration over the 2000-2020 period with respect to the 2000 value ranged from -0.83% per year for cropland sites to +1.54% per year for a wetland dominated watershed (Table 3). The Wetland category was based on one site, Snake River near Pine city, MN, receiving drained water from 2563.7 km^2 of which 34.8% is wetlands, 34.1% forest, 15.8% pastures, and only 8.8% is dedicated to cultivated crops (USGS, 2019). The FN concentration for the beginning of the trends periods at this site was 0.195 mg/l, a relatively small value compared to the average (2.8 mg/l) and the maximum (14.09 mg/l) values for all 217 trend sites. Although this site had an increase of the FN concentration, the large magnitude of the relative change with respect to the 2000 value was mainly caused by the small initial FN concentration. A substantial amount of literature has reported the property of wetlands in reducing nitrate concentration and loads (Hansen et al., 2018, Lin et al., 2008, Cheng et al., 2020, Jasper et al., 2014) hence result for this unique site is not representative of this land cover and should be further investigated. Minimum and maximum rates of change of FN nitrate-N concentration over the 2000-2020 period with respect to the 2000 for individual sites ranged between -4.65% and 4.5% per year, occurring at sites with basins dominated by cultivated cropland and forest land covers, respectively.

Mean rates of annual nitrate-N yield changes ranged from -183 kg N/km^2-yr for cultivated cropland sites to +143 kg N/km^2-yr for urban sites. Minimum and maximum rates of change ranged from -1,379 to 975 kg/km^2-yr, occurring at sites with basins dominated by cultivated and urban land covers, respectively.

5.3. Nutrient Supply and Streamflow Trend Components

The nutrient supply component was the dominant factor at 79% of sites versus 11.5% of sites where streamflow component was the dominant factor in FN concentration changes (Figure 4, Figure A- 2). Most sites having a likely decreasing concentration trend had a dominance of the nutrient component. The change in the streamflow component was greater than zero for 74% of sites (Figure 4), indicating that increased flow had some role in changing nutrient loads. At some sites where the flow component was dominant, declining FN concentration (8 sites) may be the result of dilution from increased streamflow and the depletion of a limited supply (Murphy and Sprague, 2019).

The nutrient supply component of yield trends was dominant at 49.8% of sites while streamflow component was dominant at 25.3% of sites and mixed dominance was observed at the remaining 24.9% of sites (Figure 4, Figure A- 3). The change in streamflow component was greater than zero for 94% of sites. For many sites, there was evidence of declining FN nitrate yield despite an increased streamflow component (Q2 in Figure 4 and Table 4). These sites were predominantly found in the nutrient supply dominance zone (Figure 4). Only 2 sites with flow dominance had a likely decrease on yield trends, while 51 sites had a likely increasing trend. Our results agree with those reported by Murphy and Sprague (2019) for the contiguous U.S. in which the management component (equivalent to our nutrient supply component) was the dominant component for the majority of sites, with some more importance in the yield trends than in the concentration trends.

Table 3. Mean, maximum and minimum nitrate-N trends for dominant land uses.

	% change/year Total trend concent. (%/year)			*% change/year NS trend comp. concent (%/year)*			*% change/year SF trend comp.concent. (%/year)*			*abs. change/year Total trend Yield (kg/km²-yr)*			*abs. change/year NS trend comp. Yield (kg/km²-yr)*			*abs. change/year SF trend comp. Yield (kg/km²-yr)*		
	Mean	Max	Min	Mean	Max	Min	Mean	Max	Min	Mean	Max	Min	Mean	Max	Min	Mean	Max	Min
Varied (3)	-0.43	0.39	-2.08	-0.70	-0.05	-1.84	0.27	0.60	-0.24	92.116	220.816	5.090	-13.025	11.832	-51.653	105.141	272.469	4.345
Other (21)	0.11	3.22	-3.29	-0.01	2.63	-3.24	0.12	0.84	-0.83	1.630	48.393	-46.259	-5.292	28.041	-57.987	6.922	63.881	-10.435
Pasture (14)	-0.25	2.10	-2.16	-0.43	1.95	-2.62	0.18	0.67	-0.23	90.371	260.880	-145.698	-106.777	162.666	-513.811	197.148	421.740	33.242
Urban (9)	-0.33	1.91	-1.88	0.01	2.50	-1.72	-0.33	0.01	-0.64	142.921	974.841	-265.551	49.703	826.446	-269.659	93.218	161.764	0.000
Forest (60)	0.12	4.50	-4.03	0.08	4.37	-4.12	0.04	0.53	-0.75	8.594	404.905	-510.699	-28.636	329.698	-524.013	37.230	204.167	-13.088
Cultiv. (109)	-0.83	4.22	-4.65	-1.08	3.48	-4.73	0.25	0.99	-0.72	-182.572	902.155	-1378.930	-429.310	440.309	-2364.865	246.738	1158.301	-90.210
Wetland (1)	1.54	1.54	1.54	1.49	1.49	1.49	0.05	0.05	0.05	8.166	8.166	8.166	-1.705	-1.705	-1.705	9.871	9.871	9.871
MARB (217)	**-0.40**	**4.50**	**-4.65**	**-0.55**	**4.37**	**-4.73**	**0.15**	**0.99**	**-0.83**	**-76.104**	**974.841**	**-1378.930**	**-229.089**	**826.446**	**-2364.865**	**152.986**	**1158.301**	**-90.210**

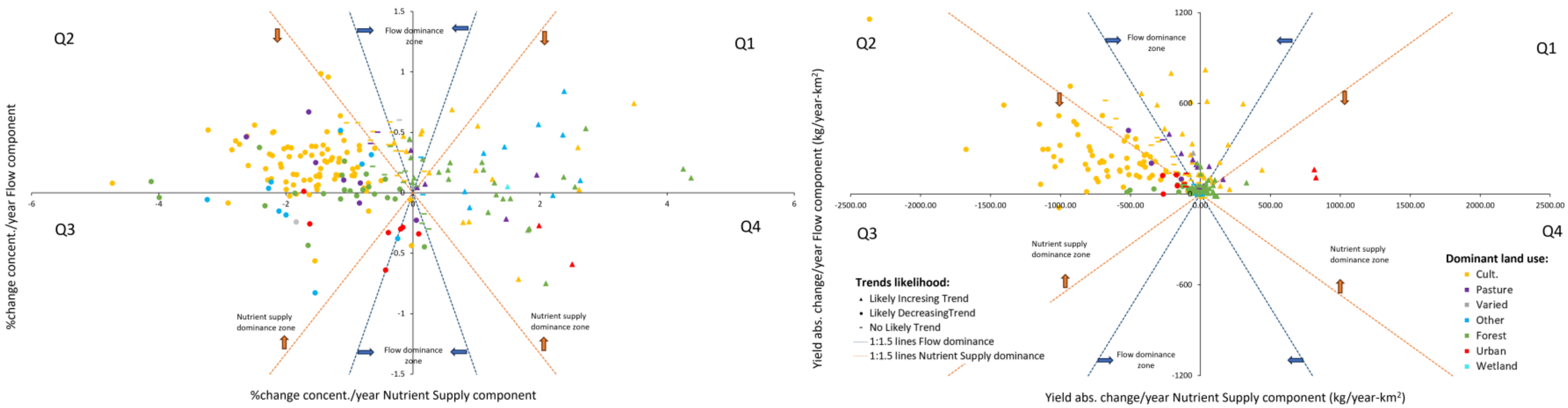


Figure 4. Nutrient Supply component vs Flow component for nitrate-N concentration and yield trends. Markers and colors represents the trend likelihood and dominant land use category, respectively.

Table 4. Percentage of sites by trend likelihood per quadrant in Figure 4

	Concentration trend categories (% of sites)				*Yield trend categories (% of sites)*			
	Likely Increas ing	**Likely decreasing**	**No likely trend**	**Cumulative Quadrant**	**Likely Increasing**	**Likely decreasing**	**No likely trend**	**Cumulative Quadrant**
Q1	16.13%	0.00%	1.38%	17.51%	23.96%	0.00%	0.00%	23.96%
Q2	3.69%	43.32%	9.68%	56.68%	13.82%	43.32%	12.90%	70.05%
Q3	0.00%	14.75%	0.00%	14.75%	0.00%	2.76%	0.00%	2.76%
Q4	7.83%	1.38%	1.84%	11.06%	1.38%	0.92%	0.92%	3.23%

5.4. Correlation Analyses

FN concentrations and yields in 2000 tended to be greatest in watersheds dominated by cultivated cropland, and there was a tendency for these high concentrations and yields to decline by 2020, with some exceptions (Figure 5). Crawford et al. (2019), analyzed nitrate trends in MARB subbasins from 2002 to 2012 and also reported a decline in yield from the basins that initially had the largest yields, but they did not incorporate land cover and trends components in their analysis as they were only considering the stationary flow normalized data, that corresponds to the nutrient supply component of our analysis. We found that, although basins with the largest 2000 concentration and yield values experienced the largest reductions in the nutrient supply component for yield trends ($r=-0.6$ and -0.77, respectively), these basins also had the largest increases in the streamflow trend component ($r=0.71$ and 0.7, respectively) (Figure 6). Increases on the streamflow component were buffered by the magnitude of the nutrient supply component decreases, producing a total decreasing effect.

FN concentrations and yields in 2000 were positively correlated ($r = 0.67$ and 0.70, respectively) with the percentage of cropland land cover in 2001. Changes in total nitrate yield, nutrient supply component and streamflow component from 2000 to 2020 were statistically ($p<0.05$) correlated ($r = -0.39$ and -0.61, and $+0.57$, respectively) with cultivated cropland cover in 2001 (Figure 6). The basin area feature had no significant correlations other than a weak correlation (r=0.21) with “other” land cover, and was removed from the correlation matrix (Figure 6) to simplify the figure.

Changes in land cover from 2001 to 2019 were relatively small and mostly uncorrelated with trends in FN concentrations or yields with a few notable exceptions (Figure A- 4). Change in cultivated cropland cover from 2001 to 2019 was positively but weakly correlated with total FN nitrate yield trend (r=0.24), nutrient supply component of nitrate yield (r-0.24) trends in the streamflow component of the FN concentration trends (r=0.25).

These results suggest that while increased streamflow promoted some increase in nitrate yield, this was counteracted by a reduced nitrate supply from the landscape at many sites. The reduced nutrient supply may be due to more efficient use of N fertilizer in cultivated cropland and reduced N discharge from point sources (McIsaac et al., 2016, Ren et al., 2022, Haegele et al., 2013).

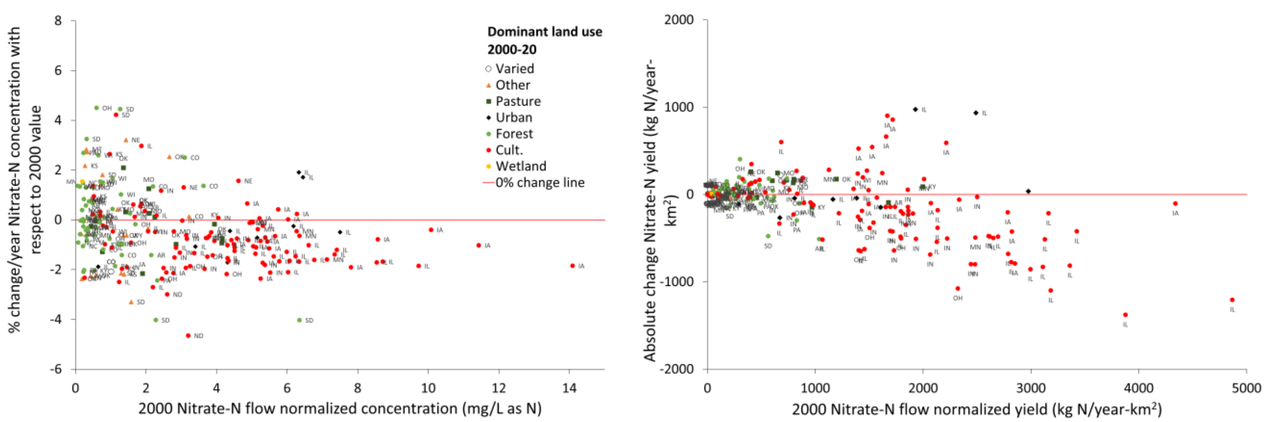


Figure 5. Scatter plots of 2000 FN values vs. 2000-20 changes for nitrate-N concentration and yield.

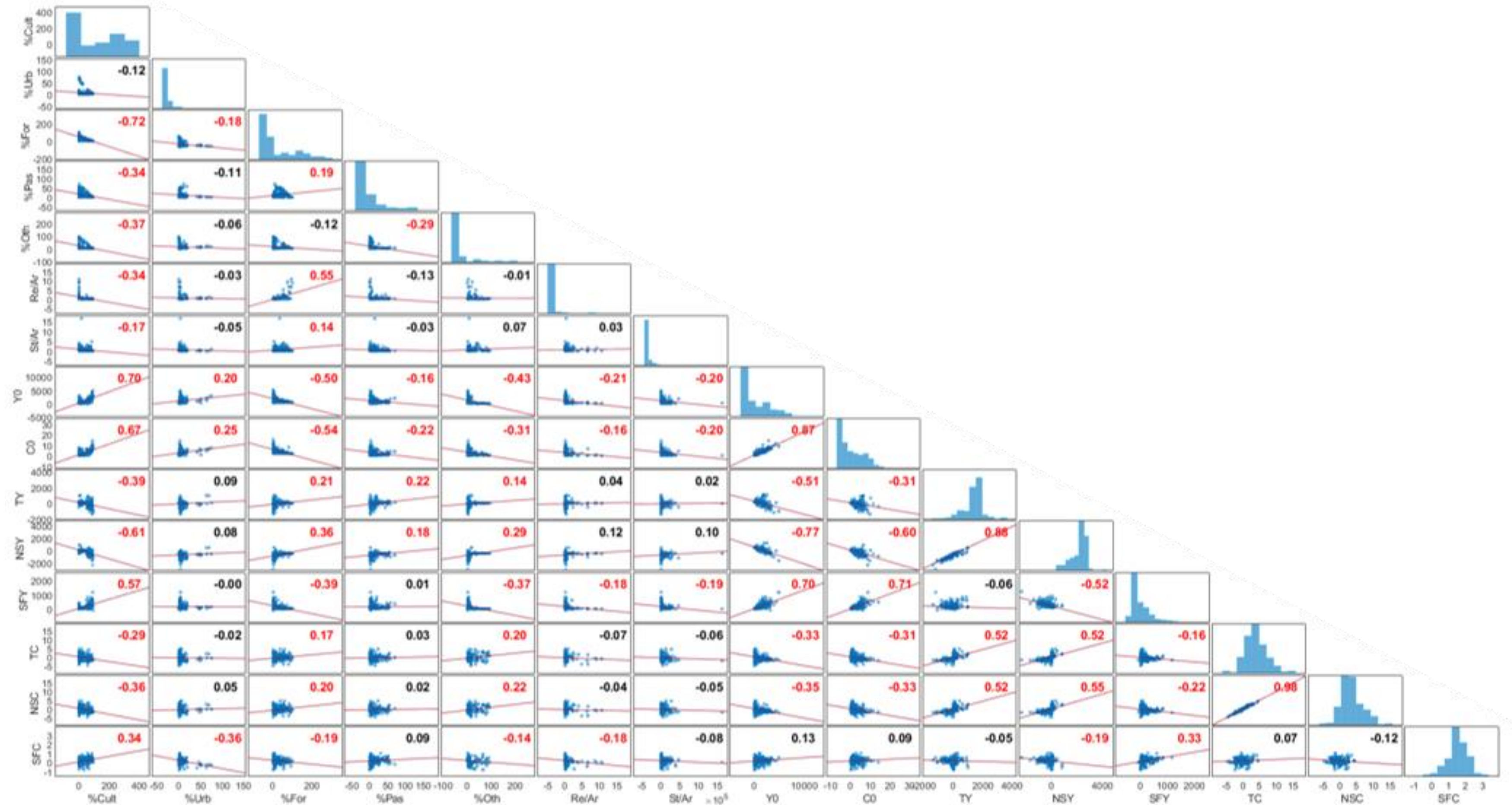


Figure 6. Pearson correlation matrix for watershed features (Table A- 1) and changes on Nitrate-N concentration (relative) and yield (absolute). Coefficient values in red are statistically significant (p<0.05).

5.5. Mississippi River and Major Tributaries

There was a high likelihood of increased ($\sigma \geq 0.9$) FN nitrate yield at the Mississippi River at Clinton, Iowa (Table 5). Several of the smaller sites upstream of Clinton also had increasing nitrate yield trends. The cluster of sites in the north central portion of the MARB that had decreased FN nitrate yield trend mostly contributed flow and load downstream of Clinton and upstream of Thebes, as does the Missouri River at Herman, MO, which had a likely increased yield trend. The increased loads at Clinton, IA and Herman, MO and perhaps some smaller tributaries appeared to offset the reduced loads from the north central cluster so that there was not a detectable trend at Thebes, IL. A similar phenomenon was reported by Crawford et al. (2019) for the 2002-2012 nutrient trends analysis at the MARB, in which load reductions at the Upper Mississippi and Ohio Rivers were offset by increasing loads and other parts of the basin, resulting in a near zero net change downstream.

There was a highly likely downward ($\sigma \leq 0.1$) trend in the Ohio River at Cannelton, IN a tributary that joins the Mississippi River downstream of Thebes, IL (Figure 7). For all four of these sites, the nutrient supply component was zero or less, while the streamflow component was greater than zero (Table 5). Only at Ohio R. at Cannelton, IN, these decreases in the nutrient supply component contributed to a highly likely reduction in the total Nitrate-N yield trend, where the percent influence of the nutrient supply component was larger than the streamflow component (Figure 7).

Table 5. Nitrate-N yield trend results for two Mississippi River mainstem sites and two major tributaries.

Location	*Yield trend*			
	Total trend (kg N/yr km²)	***Likelihood***	***Nutrient supply component (kg N/yr km²)***	***Streamflow component (kg N/yr km²)***
Mississippi R. at Clinton IA	126.3	High+	0.00	126.30
Missouri R. at Hermann MO	12.56	Likely+	-4.36	16.92
Mississippi R. at Thebes IL	17.87	Unlikely	-46.02	63.88
Ohio R. at Cannelton IN	-75.63	High-	-127.37	51.75

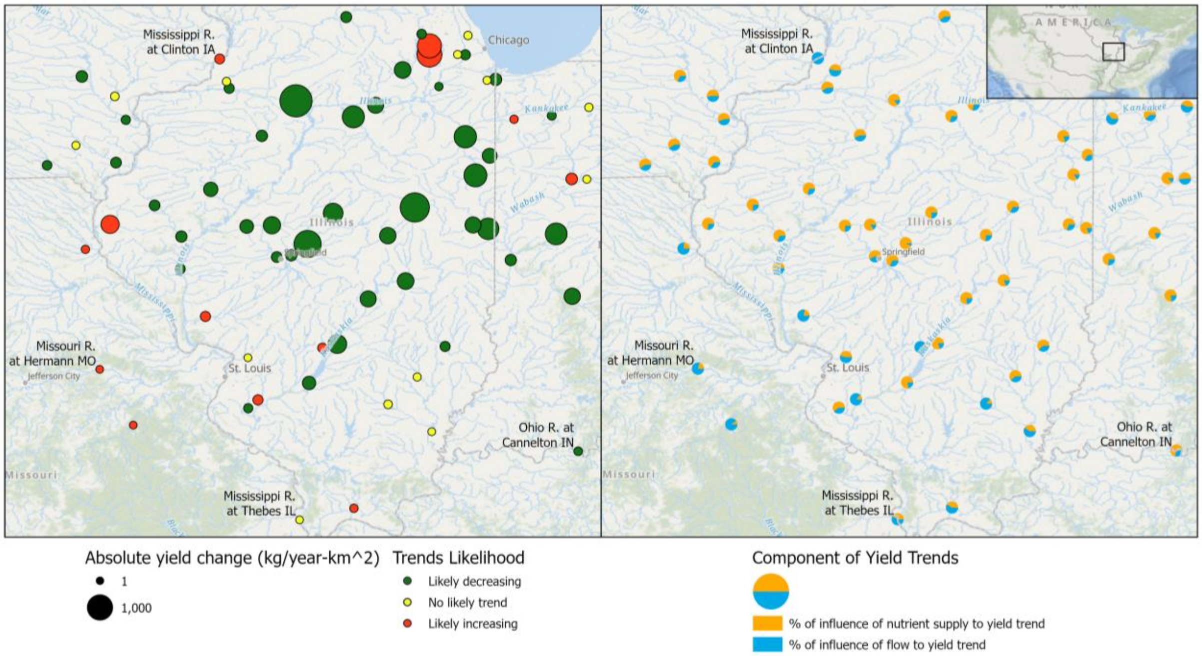


Figure 7. Location of Mississippi River mainstem and two major tributary sites.

6. Uncertainties and limitations

Flow normalized concentrations and yields are estimates of values that are hypothetically expected to occur under "normal" flow conditions based on past statistical probabilities, not on mechanistic modeling of the interactions of concentration and flow. Since flows are rarely "normal" (however normal is defined), FN concentrations and loads may differ substantially from actual concentrations and loads in a given year, which are causes of eutrophication and are the targets of nutrient reduction efforts. Flow normalization can be useful in estimating relative impacts of changes in hydrology versus other factors, but these estimates should be understood as estimates of hypothetically "normal" conditions.

Trend period selection can impact the trends magnitude and direction, since trends are estimated as the difference in concentration and yield between initial and final years. The identification of long-term flow variations and their impacts in the trends results might be impacted if the trends period is not long enough to discern random variations from non-stationary flow variability, especially at sites with extreme flow events.

Analysis of factors influencing riverine nutrient trends is limited by available data and resources. Data such as fertilizer and manure applications and crop nutrient uptake are undergoing a process of updating and refinement that is expected to be completed in 2025. These updated data should be incorporated into future studies of factors influencing riverine nutrient trends.

7. Summary and Conclusions

Screening and harmonization of nitrate concentration and associated streamflow data in the MARB resulted in 217 sites where annual FN concentrations and yields were calculated and 2000-2020 trends evaluated. Most of the watersheds dominated by cultivated cropland presented evidence of high concentrations and yields in 2000 and likely downward trends in concentration and yields over the subsequent 20 years. Many of these sites were located in the north-central portion of the basin. These reductions appear to have been offset by increased nitrate yields from the Mississippi River drainage area upstream of Clinton, Iowa and the Missouri River above Herman, MO, and other tributaries resulting in no detectable change in nitrate yield at the Mississippi River at Thebes, IL. There was a highly likely decline in nitrate yield from the Ohio River upstream of Cannelton, IN. Nitrate-N yield reductions from cropland watersheds may have been due to improved N fertilizer use efficiency, although this deserves further investigation. Causes for the increased nitrate yields upstream of Clinton, IA and in the Missouri River, basin also deserve further investigation.

For the majority of sites, the nutrient supply component was more dominant than the stream flow component for both yield and concentration. Most sites having likely decreasing concentration or yield trends had a dominance of the nutrient supply component. Some sites where the flow component was dominant, declining FN concentration (8 sites) may be the result of dilution from increased streamflow and the depletion of a limited N supply. Only 2 sites with flow dominance had a likely decrease on yield trends, while 51 sites had a likely increasing trend.

8. Data Availability

Sites information and detailed results of FN concentration and loads will be made available at the Great Lakes to Gulf Virtual Observatory web site (https://greatlakestogulf.org/nutrient-trends).

9. Acknowledgments

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10. Annexes

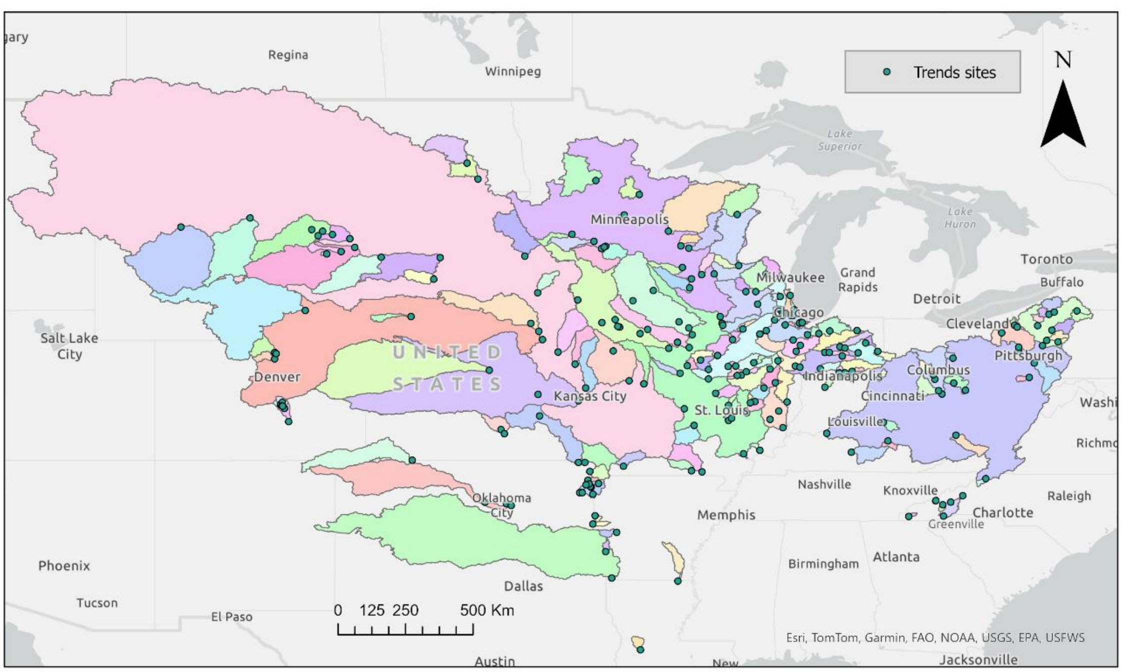

Figure A- 1. Basins and monitoring locations of the 217 n sites analyzed in this study.

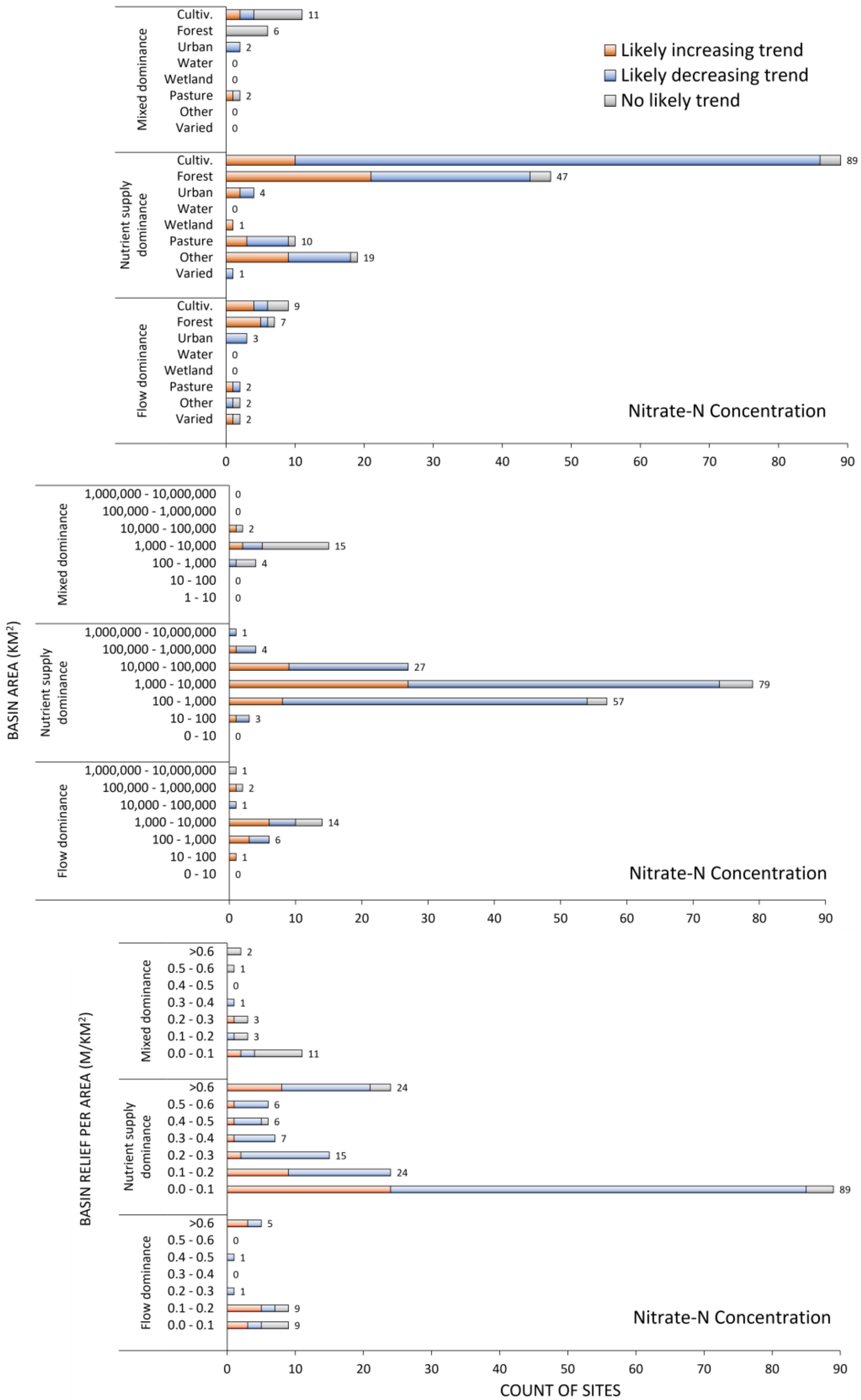

Figure A- 2. Number of trends sites per dominant concentration trend component and dominant land use, basin area and relief per area ranges. (*Varied* land use indicates a change of the dominant land use during the 2000-2020 period).

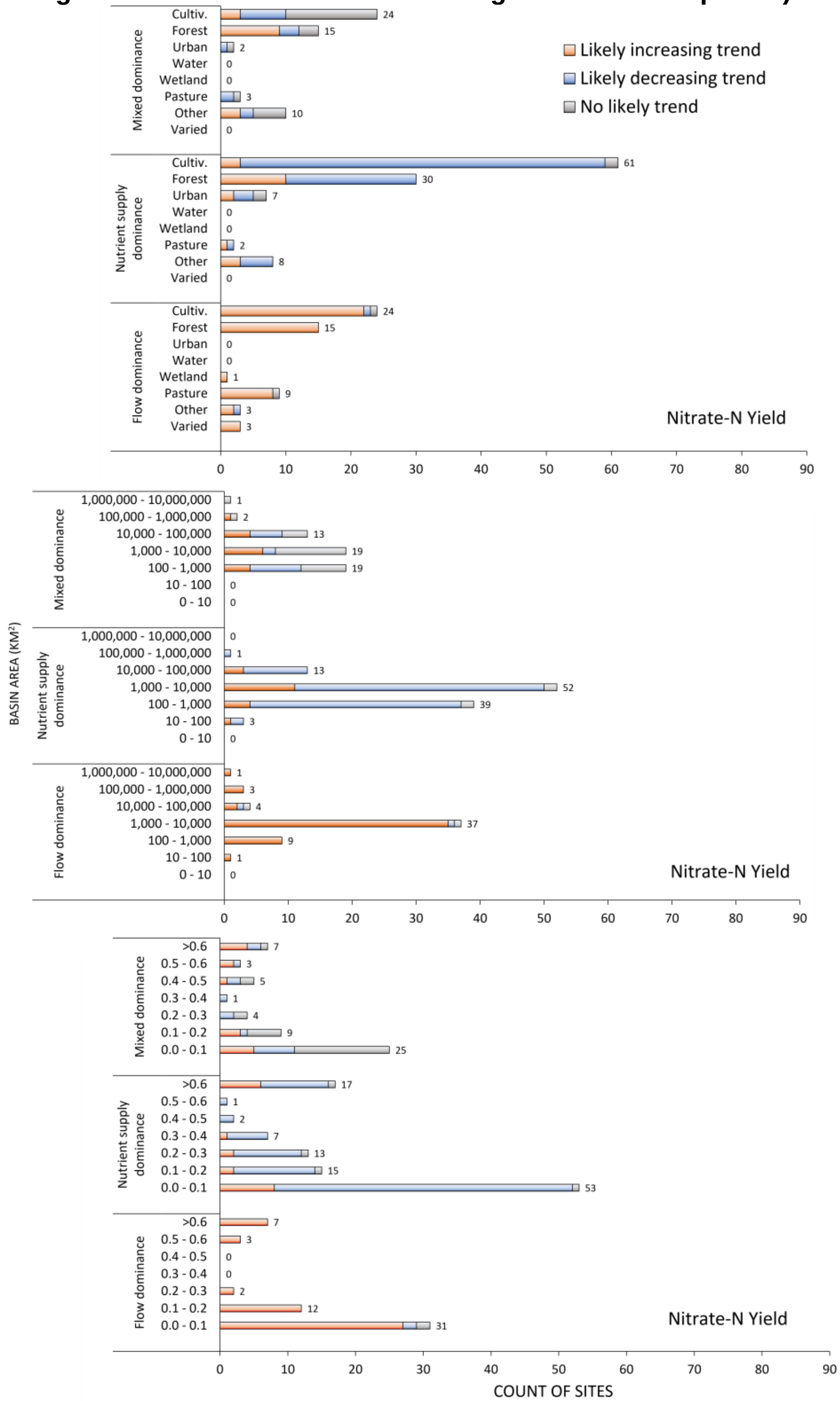

Figure A- 3. Number of trends sites per dominant yield trend component and dominant land use, basin area and relief per area ranges. (*Varied* land use indicates a change of the dominant land use during the 2000-2020 period).

Table A- 1. Abbreviations used in correlation matrices.

Variable	*Abbreviation correlation matrix*
dif2001-2019Cult. (%)	dCult
dif2001-2019Urban (%)	dUrb
dif2001-2019Forest (%)	dFor
dif2001-2019Wetland (%)	dWet
dif2001-2019Pasture (%)	dPas
dif2001-2019Other (%)	dOth
PercCu2001 (%)	%Cult
PercUr2001 (%)	%Urb
PercFo2001 (%)	%For
PercPa2001 (%)	%Pas
PercOth2001 (%)	%Oth
Area (Km^2)	Ar
Relief per area unit (m/Km^2)	Re/Ar
Upst. dam storage<2013/trend site drainage area (m^3/Km^2)	St/Ar
2000 FN Yield (kg/km^2*yr)	Y0
2000 FN Conc (mg/L)	C0
abs. change/year Total trend Yield (kg/km^2*yr)	TY
abs. change/year NS trend comp. Yield (kg/km^2*yr)	NSY
abs. change/year SF trend comp. Yield (kg/km^2*yr)	SFY
% change/year Total trend concent. (%/year)	TC
% change/year NS trend comp. concent (%/year)	NSC
% change/year SF trend comp.concent. (%/year)	SFC

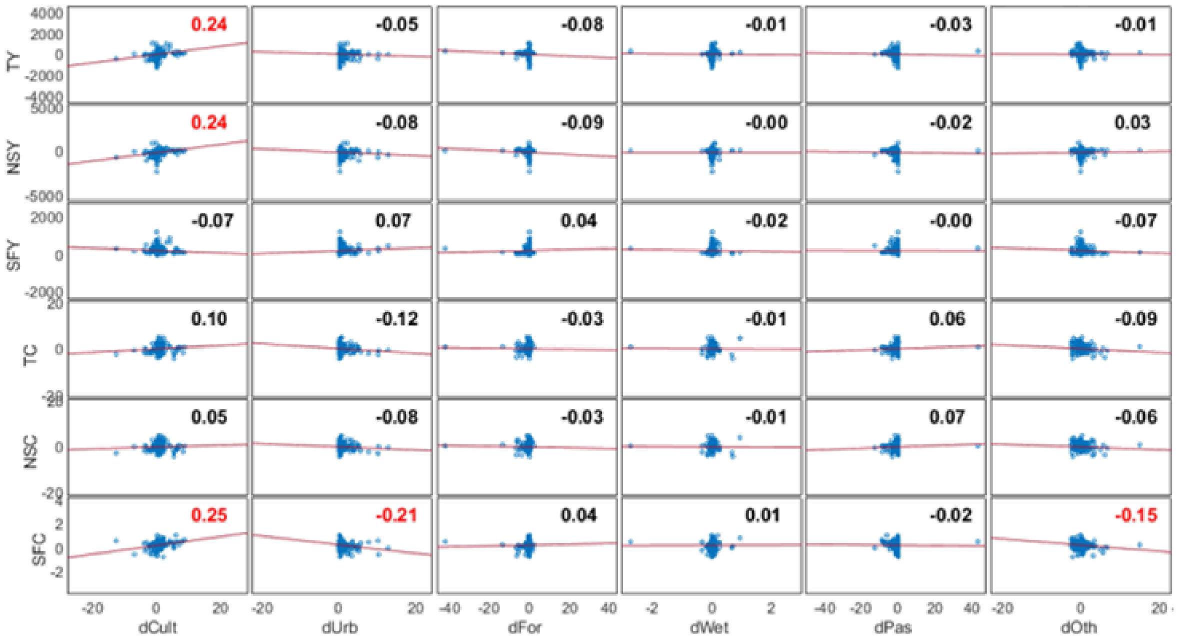

Figure A- 4. Pearson correlation matrix of percent change of land use area (Table A- 1) vs. changes in yield (absolute) and concentration (relative to 2000 values) for the 2000-20 period. Coefficient values in red are statistically significant ($p<0.05$)

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