TAMPA BAY Numeric Nutrient Criteria: Task 1 - TN and TP Concentrations

Letter Memorandum

Prepared for:



Tampa Bay Estuary Program

Prepared by:

Janicki Environmental, Inc.

Janicki Environmental, Inc.

FOREWORD

This letter memo was produced in partial fulfillment of Purchase Order #6584, TBEP Contract T-07-01 - Development of Numeric Nutrient Criteria for Tampa Bay, Task 1.

ACKNOWLEDGEMENTS

We wish to thank the partners of the Tampa Bay Estuary Program and members of the Tampa Bay Nitrogen Management Consortium for the numerous conversations providing direction and insight into concerns regarding numeric nutrient criteria establishment and appropriate methodology for developing the proposed criteria.

EXECUTIVE SUMMARY

The Tampa Bay Estuary Program (TBEP) and the Tampa Bay Nitrogen Management Consortium (TBNMC) have recommended to EPA numeric nutrient criteria for Tampa Bay as segment-specific annual total nitrogen and total phosphorus loads. EPA has noted its intention to express the numeric nutrient criteria for Tampa Bay as TN and TP concentrations. This document provides segment-specific TN and TP concentrations consistent with the TN and TP loads recommended as numeric nutrient criteria.

Establishment of numeric nutrient criteria is dependent on an understanding of the limiting nutrient within the water body of concern. For Tampa Bay, extensive data exist for evaluation of which nutrient, nitrogen or phosphorus, is limiting. Ambient water quality data strongly indicate that Tampa Bay is nitrogen limited, and this is supported by the results of nutrient addition bioassays.

Previous efforts by the TBEP have developed strong relationships between nutrient supply to Tampa Bay and resultant chlorophyll a concentrations in the bay, and between chlorophyll a concentrations and light availability for seagrasses. Thus, management actions have focused on controlling nitrogen loads to Tampa Bay, with measureable success as expressed by increases in seagrass acreage. The relationships are between nitrogen loads and chlorophyll *a*, however, not nitrogen concentrations in the bay and chlorophyll *a*.

Previous efforts to link in-bay nitrogen concentrations to chlorophyll a concentrations have not produced sufficient explanatory relationships for use in nitrogen concentration target setting. The current effort, driven by EPA's intention to express numeric nutrient criteria as in-bay concentrations, examines several methods to link nutrient concentrations to chlorophyll a concentrations and/or nutrient loads.

The final method selected for establishing proposed concentration numeric nutrient criteria for nitrogen and phosphorus was the reference period approach, based on the following rationale. Segment-specific chlorophyll a targets (values at this level or below indicate desirable conditions) have been established previously (Janicki and Wade, 1996). These targets were based on a 1992-1994 reference period. In 2001, a protocol for assessing whether the Tampa Bay segments were achieving these targets was developed (Janicki Environmental, 2001). This protocol, referred to as the Decision Matrix approach, considered the year-to-year variability in chlorophyll a concentrations and arrived at segment-specific chlorophyll a thresholds (values above this level indicate undesirable conditions). The threshold was the sum of the chlorophyll a target and 2X the standard error of the long-term chlorophyll a concentrations.

Using the same approach, the segment-specific annual geometric mean nitrogen and phosphorus concentrations of the 1992-1994 period were derived as commensurate with the chlorophyll *a* targets. These concentrations were increased by one standard deviation (as derived from 1992-2009 data) to develop the proposed concentration numeric nutrient criteria, which serve as the threshold concentrations in the bay.

The following conclusions are drawn from the analyses completed for this effort:

- Tampa Bay is nitrogen-limited as indicated by both ambient TN:TP ratios and nutrient addition bioassays.
- There is no discernable relationship between TN loadings and in-bay TN concentrations or between TP loadings and in-bay TP concentrations in any bay segment. This is not because loadings do not affect in-bay concentrations, but because various other confounding factors, for which sufficient data are not available, play a role in relationships between loadings and concentrations.
- On a monthly time scale, the relationships between either TN concentrations or TP concentrations and chlorophyll a concentrations do not explain a significant proportion of the variability in the chlorophyll a concentrations to support development of concentration-based numeric nutrient criteria in any bay segment.
- There are differences in TN concentrations and TP concentrations, particularly in Hillsborough Bay and Middle Tampa Bay, in those years when the chlorophyll a concentration thresholds are met when compared to those observed in years when the chlorophyll a concentration thresholds are not met. However, there is a great deal of variability within the data obtained in either group of years and these differences are not recommended as the basis for the establishment of concentration-based numeric nutrient criteria for Tampa Bay.
- The reference period approach is recommended for the establishment of concentration-based TN and TP criteria for Tampa Bay. The segment-specific annual geometric mean nitrogen and phosphorus concentrations of the 1992-1994 period were increased by one standard deviation (as derived from 1992-2009 data) to develop the proposed concentration numeric nutrient criteria. These criteria are:

-	Old Tampa Bay	TN = 0.93 mg/L	TP = 0.31 mg/L
-	Hillsborough Bay	TN = 1.01 mg/L	TP = 0.45 mg/L
-	Middle Tampa Bay	TN = 0.87 mg/L	TP = 0.29 mg/L
-	Lower Tampa Bay	TN = 0.74 mg/L	TP = 0.10 mg/L.

The criteria referenced above should be assessed as an annual geometric mean from long-term, monthly water quality monitoring stations currently used in the state's chlorophyll a threshold assessments under the Tampa Bay Reasonable Assurance determination. The assessment of TN and TP concentrations attainment should only occur when chlorophyll a thresholds are exceeded within a bay segment, and should coordinate with current regulatory assessments under the FDEP RA determination and EPA TMDL for TN loads in Tampa Bay. Further, compliance assessments should be conducted over five-year time frames, with no more than two consecutive years being greater than these established criteria if chlorophyll a thresholds are also exceeded during the same time period. This approach is analogous to the chlorophyll a threshold assessments currently being conducted under the regulatory requirements for the FDEP RA determination and EPA TMDL for Tampa Bay.

1.0 Introduction and Objective

The Tampa Bay Estuary Program (TBEP) and the Tampa Bay Nitrogen Management Consortium (TBNMC) have recommended numeric nutrient criteria to U.S. Environmental Protection Agency (EPA) for Tampa Bay (TBNMC, 2010). The criteria, as proposed to EPA, are segment-specific (Figure 1) and are expressed as annual total nitrogen (TN) and total phosphorus (TP) loads. These TN and TP loads are those for the reference period of 1992-1994, as discussed in the March 8, 2010 comments to EPA. However, EPA has informed the TBEP that it intends to express the numeric nutrient criteria for Tampa Bay as TN and TP concentrations.

The objective of this task is to develop segment-specific TN and TP concentrations consistent with the TN and TP loads recommended as numeric nutrient criteria by the TBEP and TBNMC. This is in keeping with recognition of the importance of maintaining consistency with existing management goals and specifically with the recent load allocations to comply with the existing TMDL for Tampa Bay.

The following provides a discussion of nutrient limitation, description of the analyses completed and the results of each analysis, and the recommended TN and TP numeric nutrient criteria, expressed as concentrations, for each of the four mainstem bay segments.

2.0 Nutrient Limitation

The establishment of numeric nutrient criteria depends upon knowledge of the nutrient most likely limiting in the waterbodies of concern. Three major factors control whether nitrogen or phosphorus is more likely to be limiting (NRC, 2000):

- the N:P ratio in external nutrient inputs;
- the preferential loss from the photic zone of nitrogen or phosphorus due to biogeochemical processes such as denitrification, sedimentation, or absorption of phosphorus; and
- the amount of nitrogen fixation.

Marine systems, including estuaries, are generally considered nitrogen limited (Thomas, 1970a,b; Ryther and Dunstan, 1971; Boynton et al., 1982; Smith, 1984; Howarth, 1988, 2008; Howarth et al., 1988a,b; Nixon et al., 1996; Howarth and Marino, 2006; Chapra, 1997; National Research Council, 2000;), although there may be times and locations when phosphorus limitation may occur (Conley, 2000; Conley et al., 2009; Malone et al., 1996).

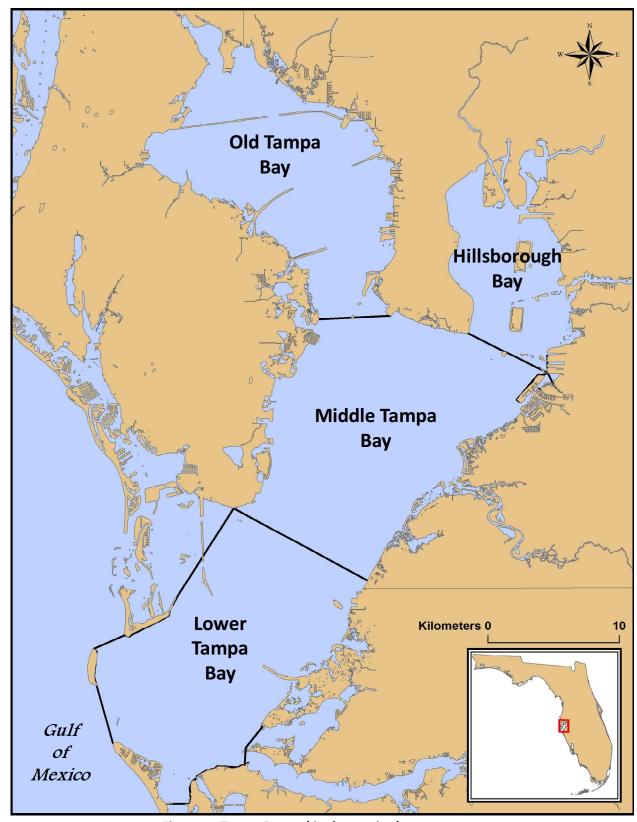


Figure 1. Tampa Bay and its four major bay segments.

Since nitrogen is considered the most likely limiting nutrient in estuarine systems, it has been identified as the primary nutrient of concern in estuarine ecosystems nationwide (Smith, 1984; NRC, 1993). As noted in Correll (1999), however, since estuaries are part of the transition zone between the open ocean and the phosphorus supplied from the land, it is possible that both phosphorus and nitrogen may be limiting in estuaries, dependent upon the time of year, location in the estuary, and nutrient supplies. Ryther and Dunstan (1971) noted the change from phosphorus limitation in freshwaters to nitrogen limitation in near-shore marine waters, although Hecky and Kilham (1988) indicated that the extent and severity of marine nitrogen limitation has not been conclusively determined, with other studies reporting estuarine phosphorus limitation in the spring and nitrogen limitation in the summer and fall (Fisher et al., 1992; Lee et al., 1996). Depending upon the relative rates of nitrogen and phosphorus supply, the limitation has been found to shift between nitrogen and phosphorus in coastal lagoons in the northeastern US (Taylor et al., 1995). It has also been documented that residence times play a significant role in determining the estuarine responses to nutrient loads ((Monson et al., 2002; Hagy et al., 2000; Borsuck et al., 2004; Boynton and Kemp, 2008).

2.1 Methods to Determine Limitation

There are two general methods that have been used to define which nutrient is limiting in a water body. They include:

- a method that depends upon ambient water quality data collected over a wide range of environmental conditions, and
- a method that involves experimental manipulation of nutrient conditions, either in the laboratory or *in situ*.

2.1.1 Ambient Water Quality Data Methods

This method depends upon a metric typically used to evaluate nutrient limitation: the nitrogen to phosphorus ratio (N:P ratio). The N:P ratio indicative of balanced conditions is typically taken to be 16:1 (molar), based on the work of Redfield (1934, 1958). This N:P ratio was based on the elemental composition of algae, under both laboratory and natural conditions. When N:P ratios are greater than 16:1 in a system, this is indicative of phosphorus limitation. However, there may be considerable variation in this ratio within an algal culture, dependent upon cell division status, light conditions, and precedent conditions (Correll, 1999; Correll and Tolbert, 1962; Terry et al., 1985).

Molar ratios of N:P are easily determined from water quality monitoring data. In freshwater systems, the N:P ratio is usually higher than 16:1, indicating that phosphorus is usually most limiting to primary production in these ecosystems (Schindler, 1977; Elser et al., 2007). This becomes evident by examining the concentrations of the forms of nitrogen and phosphorus that are available for algal uptake. Little if any dissolved inorganic phosphorus (DIP) is generally found in relatively productive freshwaters while measurable concentrations of dissolved inorganic nitrogen (DIN) remain.

Most marine systems are nitrogen limited because there are relatively low concentrations of dissolved inorganic nitrogen compared to dissolved phosphorus. Since Redfield's observations were published, research has shown that ratios from 10:1 to 20:1 for N:P are typically found in

estuaries (Parsons et al., 1984). Howarth (1988) observed that the correlation between nitrogen and the primary production was better for estuaries that received nutrient concentrations with smaller N:P ratios than the one studied by Redfield. Several studies have led to the conclusion that estuaries receiving nutrient concentrations with high N:P ratios were limited by phosphorus and only those with low ratios are limited by nitrogen (Boynton et al., 1982). Boynton et al. (1982) and Howarth (1988) compiled data on the ratio of inorganic nitrogen to phosphorus in a variety of estuaries. Of the 27 studied by Howarth, 22 had N:P ratios below the Redfield ratio and may have been nitrogen limited. Because phytoplankton can assimilate some organic nutrient forms and all forms are relatively labile, it is useful to examine the ratio of total nutrient concentrations (TN:TP).

Reductions of nutrient levels in a water body will usually result in reduction in algal growth. Reducing phosphorus, however, will have no effect unless the reduction results in an N:P ratio greater than 16:1. Phosphorus would then become the limiting nutrient. In contrast, a reduction of nitrogen concentrations will result in a reduction of primary productivity when the ratio is less than 16:1. There are exceptions to this general rule. Some coastal areas are phosphorus limited due to strict phosphorus control measures or natural conditions and some freshwaters are nitrogen limited due to natural sources of phosphorus.

2.1.2 Experimental Methods

Experimental manipulation of nutrient conditions, either in the lab or *in situ*, typically involves nitrogen and phosphorus additions to either a test alga or a phytoplankton assemblage singularly and in combination. The responses to the additions determine the limiting nutrient. If growth is found only during nitrogen addition, nitrogen-limitation is indicated. Conversely, if growth is found only during phosphorus addition, phosphorus-limitation is indicated.

In situ methods have included:

- limnocorrals or bags in which nutrient additions are made and resultant growth responses are measured (Shapiro, 1980; Lynch and Shapiro, 1981; Havens and DeCosta, 1986; Perez et al., 1994);
- mesocosm studies in which water is collected and placed in separate containers or enclosures for application of separate treatments over multiple day time scales (Oviatt et al., 1986; Taylor et al., 1995); and
- whole-lake studies performed on entire lakes or portions of lakes separated by curtains (Schindler, 1974, 1975).

In Florida, as part of its TMDL process, the Florida Department of Environmental Protection (FDEP) attempts to identify the limiting nutrient(s) in impaired waterbodies. The TMDL for a specific waterbody specifies the maximum amount of the limiting nutrient that may enter the waterbody, with this limitation being defined with the aim of improving water quality. If the N:P ratio does not clearly suggest the limiting nutrient, TMDLs for both nitrogen and phosphorus are typically defined. The primary method for determining the limiting nutrient employed by the FDEP is use of existing water quality data to derive ambient N:P ratios, but more complicated methods, including field tests and laboratory algal growth potential bioassays, have been employed. Per FDEP guidelines, receiving waters with ratios less than 10:1 (molar) are considered nitrogen limited, ratios of greater than 30:1 (molar) indicate phosphorus limitation, and ratios of 10-30:1 (molar) indicate colimitation (FDEP, 2002).

2.2 Confounding Factors

Determination of the limiting nutrient based solely on N:P ratios estimated from water quality data or from experimental uptake rates should be performed with consideration of potentially confounding effects. Algal cell interior N:P ratios and uptake rates may vary due to:

- cell division status (Correll and Tolbert, 1962),
- light intensity or light quality (Wynne and Rhee, 1986),
- light and temperature (Jahnke et ., 1986), and
- P deprivation and then subsequent availability (Sicko-Goad and Jensen, 1976).

Nutrient limitation in freshwaters, which are typically considered to be phosphorus limited, can vary seasonally. Summer nitrogen limitation in lakes can occur when photic zone inorganic nutrients are low (Elser et al., 1990). It has also been demonstrated that some estuaries show seasonal shifts in limitation (D'Elia et al., 1986; McComb et al., 1981; Conley, 2000). The best available information should be used to determine the limiting nutrient of a system before management decisions are made with the objective of improved water quality via nutrient load control.

2.3 Nutrient Limitation in Tampa Bay

Nutrient limitation in Tampa Bay has been examined using both the N:P ratio method and nutrient addition bioassays.

2.3.1 TN:TP Ratios in Tampa Bay

The average TN:TP ratios for the segments of the TBEP, both by weight and molar, were determined based on ambient water quality data, and are presented in Table 1 for the period 1981 to 2009. The ratios were calculated by first calculating the monthly ratio for each segment based on data collected by the Environmental Protection Commission of Hillsborough County (EPCHC). The mean value of these monthly values within a year was calculated and the mean of these annual values was calculated.

Table 1. Annual mean TN and TP concentrations and TN:TP in TBEP segments (1981-2009).					
Bay Segment	TN (mg/L)	TP (mg/L)	TN:TP (Weight)	TN:TP (Molar)	
Old Tampa Bay	0.71	0.27	3.5	7.7	
Hillsborough Bay	0.79	0.45	2.1	4.6	
Middle Tampa Bay	0.63	0.27	2.9	6.3	
Lower Tampa Bay	0.48	0.13	4.8	10.5	

All segments except Lower Tampa Bay have molar N:P ratios less than 10:1, while that for Lower Tampa Bay is just slightly greater than 10:1. According to the FDEP guidelines (FDEP, 2002), all segments would therefore be considered nitrogen-limited. While nitrogen limitation is indicated in all segments, the nutrient that is most limiting can vary seasonally (Malone et al., 1996; Conley et

al., 2009), so that areas that are generally nitrogen limited may by phosphorus-limited at times. In addition to nutrient limitation, phytoplankton growth may also be light-limited during certain parts of the year (Pennock and Sharp, 1994).

Seasonal variation in nutrient limitation has been observed in other waterbodies (Fisher et al., 1992; Lee et al., 1996; Malone et al, 1996; Conley et al., 2009). Season-specific TN:TP ratios were also estimated based on the 1981-2009 data (Table 2). These estimates continue to support the conclusion that Tampa Bay is nitrogen-limited.

Table 2. Seasonal mean TN:TP ratios in TBEP segments (1981- 2009).				
	Dry Season		Wet Season	
Bay Segment	TN:TP	TN:TP	TN:TP	TN:TP
	(Weight)	(Molar)	(Weight)	(Molar)
Old Tampa Bay	3.6	8.0	3.2	7.1
Hillsborough Bay	2.2	4.9	1.9	4.2
Middle Tampa Bay	3.0	6.6	2.6	5.7
Lower Tampa Bay	5.1	11.3	4.2	9.2

2.3.2 Nutrient Addition Bioassays in Tampa Bay

The City of Tampa Bay Study Group routinely performed bioassays throughout Tampa Bay during 1993-2009 as part of the evaluation of the effects of discharge from the H.F. Curren wastewater facility (Johansson, 2009 – Attachment 1). Bioassays in late winter and late summer have been conducted for most of this period. The results of these 152 bioassays have supported that nitrogen is the primary limiting nutrient in the bay, with no results showing phosphorus as the limiting nutrient in the bay, including within that portion of the bay which receives the wastewater effluent. Therefore, the discharge phosphorus limitation is not required for this facility.

It is concluded that the four mainstem bay segments in Tampa Bay are nitrogen-limited. This conclusion contributed to the development of TN loading targets for Tampa Bay (Janicki and Wade, 1996), without consideration of TP loading targets in the original target setting effort. Since EPA is considering developing both TN and TP numeric nutrient criteria for Tampa Bay, the proposed numeric nutrient criteria expressed as TP loads were based on the same time period used to develop the proposed TN loading numeric nutrient criteria (comments submitted to EPA (TBNMC, 2010). Table 3 presents the recommended protective nutrient loads established by the TBNMC and proposed to EPA.

Table 3. Protective nutrient loads for the Tampa Bay estuary established by the Tampa Bay Nitrogen Management Consortium, and accepted through separate administrative action by FDEP (acceptance of the 2002 RA, 2007 RA Update & 2009 RA Addendum) and EPA (establishment of the 1998 federally-recognized TMDL for Tampa Bay).

Bay Segment	Tampa Bay NMC Proposed Alternative Total Nitrogen Load expressed as tons/year	Tampa Bay NMC Proposed Total Phosphorus Load expressed as tons/year
Old Tampa Bay	486	104
Hillsborough Bay	1 <i>,</i> 451	1,093
Middle Tampa Bay	799	140
Lower Tampa Bay	349	52
Remainder of Lower Tampa Bay	629	112

3.0 Tampa Bay TN and TP Criteria: Analyses and Results

Multiple analyses were completed in the evaluation of potential TN and TP criteria expressed as inbay concentrations. These include:

- examination of the relationships between TN and TP loadings to in-bay TN and TP concentrations, respectively;
- examination of relationships between monthly TN and TP concentrations with chlorophyll a concentrations;
- examination of relationships between annual TN and TP concentrations with chlorophyll a concentrations; and
- application of a reference period approach to establishing TN and TP concentration-based criteria.

The data used in these analyses are defined in Attachment 2. The following describes these analyses and the results obtained.

3.1 Evaluation of Relationships Between In-bay TN and TP Concentrations and TN and TP Loads

Since the current proposed TN and TP criteria are expressed as loads, the simplest method to propose criteria expressed as in-bay concentrations would be based on the potential relationships between in-bay TN and TP concentrations and TN and TP loads delivered to each segment. All four mainstem segments have annual TMDL TN loads recognized by both EPA and DEP. If significant relationships are found between the nutrient loads and their respective in-bay concentrations, then the proposed numeric nutrient criteria loads could be expressed as concentrations.

Monthly segment-specific TN and TP concentrations were merged with monthly segment-specific TN and TP loads resulting in a dataset of monthly values of TN and TP concentrations and loads. Plots of these data were inspected, with TN and TP concentrations as functions of TN and TP loads, respectively, including various lag and cumulative load effects. Graphical results for each segment

are provided in Attachment 3 (TN) and Attachment 4 (TP). The relationships examined are between the in-bay nutrient concentrations and the following:

- Current months load,
- Previous month load,
- Cumulative loads over current and previous month, and
- Cumulative loads over current and two previous months.

The graphical representations of the relationships were then used to guide evaluation of relationships between TN and TP concentrations and loads that may explain large proportions of the monthly variation in concentrations.

No relationships were found between TN concentrations and potential TN load explanatory variables that explained more than 20% of the variation in TN concentrations in Hillsborough Bay or more than 10% of the variation in concentrations in the other three bay segments (Table 4). In Old Tampa Bay, the natural log-transformed three-month cumulative TN load explained only 7% of the variation in in-bay TN concentrations. In Hillsborough Bay, the natural log-transformed twomonth cumulative TN load explained 33% of the variation in in-bay TN concentrations. In Middle Tampa Bay, 10% of the variation in in-bay TN concentrations was explained by the natural logtransformed three-month cumulative TN load, and 20% of the variation in in-bay TN concentrations in Lower Tampa Bay was explained by the natural log-transformed cumulative threemonth TN load. This suggests that there are some in-bay processes (e.g., sedimentation, denitrification, and transport within the bay and exchange with the Gulf of Mexico) that affect the relationship between the TN loads and the resultant in-bay TN concentrations in Tampa Bay. The relationships between TP concentrations and potential TP loading explanatory variables that were found varied appreciably among bay segments (Table 3). In Old Tampa Bay, the natural logtransformed three-month cumulative TP load explained 44% of the variation in TP concentrations. In Hillsborough Bay, the natural log-transformed current month TP load explained 33% of the variation in in-bay TP concentrations. In Middle Tampa Bay, 47% of the variation in TP concentrations was explained by the natural log-transformed three-month cumulative TP load, and 20% of the variation in TP concentrations in Lower Tampa Bay was explained by the natural logtransformed cumulative three-month TP load.

The results of these analyses do not provide adequate evidence to support recommendations for TN and TP concentration criteria based on the relationships between the in-bay nutrient concentrations and the nutrient loads to those segments.

Table 4. Best-fit regressions of TN and TP concentrations on TN and TP loads, respectively.					
Segment – Variable	Regression		r ²		
Old Tampa Bay – [TN]	[TN] = 0.19 + 0.112 * In Cumulative 3-Month TN Load	< 0.0001	0.07		
Old Tampa Bay – [TP]	[TP] = -0.32 + 0.15 *In Cumulative 3-Month TP Load	< 0.0001	0.44		
Hillsborough Bay – [TN]	[TN] = -0.25 + 0.189 * In Cumulative 2-Month TN Load	< 0.0001	0.20		
Hillsborough Bay – [TP]	[TP] = 0.32 + 0.0008 * In Current Month TP Load	< 0.0001	0.33		
Middle Tampa Bay – [TN]	[TN] = 0.5 + 0.0005 *In Cumulative 3-Month TN Load	< 0.0001	0.10		
Middle Tampa Bay – [TP]	[TP] = -0.08 + 0.089 * In Cumulative 3-Month TP Load	< 0.0001	0.47		
Lower Tampa Bay – [TN]	[TN] = 0.3 + 0.036 *In Cumulative 3-Month TN Load	< 0.0141	0.02		
Lower Tampa Bay – [TP] $[TP] = 0.1 + 0.0005 * ln Cumulative 3-Month TP Load < 0.0005 * ln Cumulative 3 + Month TP Load$		< 0.0001	0.20		

3.2 Evaluation of Relationships Between Chlorophyll a and TN and TP Concentrations

The second data analysis approach examined the potential relationships between chlorophyll a concentrations and either TN or TP concentrations in each bay segment. Chlorophyll a thresholds have been established by the TBEP as part of the nitrogen management plan for the bay. If significant relationships are found, then the chlorophyll a thresholds could be used to determine the corresponding nutrient concentrations for use as numeric nutrient criteria. There are two temporal scales that can be examined – monthly and annual.

3.2.1 Evaluation of Relationships Between Monthly Chlorophyll *a* and TN and TP Concentrations

Initially, monthly segment-specific chlorophyll a and nutrient (TN and TP) concentrations were plotted. A series of variables based on the ambient TN and TP concentrations, including various lag concentrations, was examined. Graphical results are provided in Attachments 5 (TN) and 6 (TP) for each segment as scatter plots of chlorophyll a and the following:

- Current month TN and TP concentration,
- Previous month TN and TP concentration,
- Average of current and previous months TN and TP concentrations, and
- Average of current and two previous months TN and TP concentrations,

The graphical representations of the relationships were then used to guide evaluation of relationships between chlorophyll *a* and TN and TP concentrations that may explain the monthly variation in chlorophyll *a* concentrations.

No relationships were found between chlorophyll a and TN concentrations that explained more than 24% of the variation in chlorophyll a (maximum $r^2 = 0.24$) (Table 5). In Old Tampa Bay, the mean two-month TN concentration explained 12% of the variation in in-bay chlorophyll a concentrations. In Hillsborough Bay, the same-month TN concentration explained 33% of the variation in in-bay chlorophyll a concentrations. In Middle Tampa Bay, 17% of the variation in in-bay chlorophyll a concentrations was explained by the same-month TN concentration, and only 6% of the variation in in-bay chlorophyll a concentrations in Lower Tampa Bay was explained by the same-month TN concentration.

No relationships were found between chlorophyll *a* and TP concentrations that explained more than 27% of the variation in chlorophyll *a* concentrations (Table 5). In Old Tampa Bay, the samemonth TP concentration explained 15% of the variation in in-bay chlorophyll *a* concentrations. In Hillsborough Bay, the mean two-month TP concentration explained 17% of the variation in in-bay chlorophyll *a* concentrations. In Middle Tampa Bay and Lower Tampa, the same-month TP concentration explained 27% and 12%,respectively, of the variation in in-bay chlorophyll *a* concentrations.

Table 5. Best-fit regressions of monthly chlorophyll a concentrations on TN and TP						
concentrations.	concentrations.					
Segment – Variable	Regression	p > F	r ²			
Old Tampa Bay – TN	chl $a = 3.78 + 7.34$ *Mean 2-month TN Concentration	< 0.0001	0.12			
Old Tampa Bay – TP	chl $a = 5.51 + 14.94*$ Mean TP Concentration	< 0.0001	0.15			
Hillsborough Bay – TN	chl $a = 1.22 + 15.7*$ Mean TN Concentration	< 0.0001	0.24			
Hillsborough Bay – TP	chl $a = 6.82 + 15.65*$ Mean 2-month TP Concentration	< 0.0001	0.17			
Middle Tampa Bay – TN	chl $a = 2.75 + 7.45*$ Mean TN Concentration	< 0.0001	0.17			
Middle Tampa Bay – TP $chl a = 3.45 + 15.59*Mean TP Concentration$ $< 0.0001 0.27$						
Lower Tampa Bay – TN	chl $a = 3.02 + 2.9*$ Mean TN Concentration	< 0.0003	0.06			
Lower Tampa Bay – TP	chl $a = 2.97 + 10.22*Mean TP Concentration$	< 0.0001	0.12			

The results of these analyses do not provide adequate evidence to support recommendations for TN and TP concentration criteria based on the relationships between the in-bay chlorophyll *a* concentrations and the in-bay TN and TP concentrations in these segments.

3.2.2 Evaluation of Relationships Between Annual Chlorophyll a and TN and TP Concentrations

The relationships between annual mean chlorophyll a and annual mean TN and TP concentrations were examined as a potential means of developing nutrient criteria. The annual mean TN and TP concentrations were classified according to whether the annual chlorophyll a threshold in a given segment was met or not. Based on this classification, the TN and TP concentrations for those years in which the threshold was met could be compared to those TN and TP concentrations for those years when the threshold was exceeded (Figures 2-9).

The results of this analysis indicate that on an annual basis, there are differences in the in-bay TN concentrations during years in which the threshold chlorophyll a targets are met and those in which they are not met. This is particularly the case in Hillsborough Bay and Middle Tampa Bay. Similar differences were found between the TP concentrations in those years in which the threshold chlorophyll a targets are met and those in which they are not met. However, these differences are not as discrete as seen in the TN concentrations.

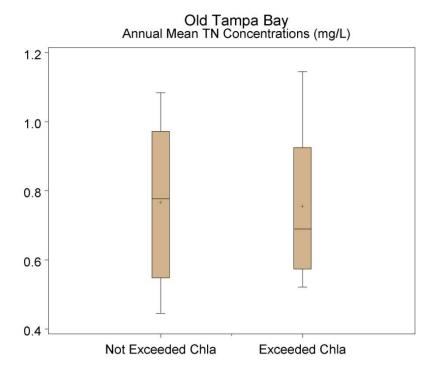


Figure 2. Comparison of TN concentrations in Old Tampa Bay in those years when the chlorophyll *a* thresholds were exceeded and those years when the chlorophyll *a* thresholds were not exceeded.

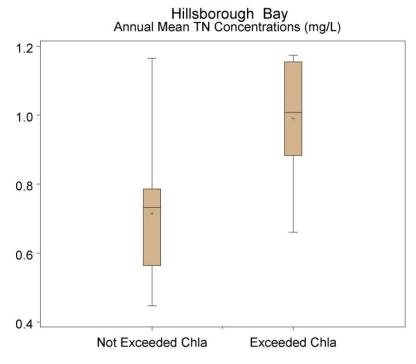


Figure 3. Comparison of TN concentrations in Hillsborough Bay in those years when the chlorophyll *a* thresholds were exceeded and those years when the chlorophyll *a* thresholds were not exceeded.

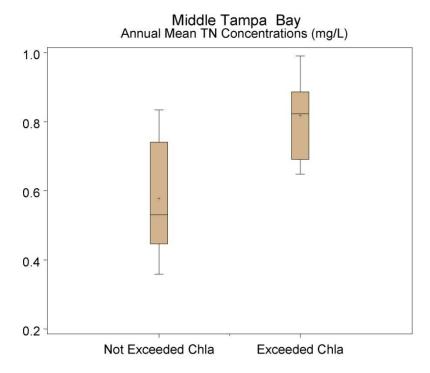


Figure 4. Comparison of TN concentrations in Middle Tampa Bay in those years when the chlorophyll *a* thresholds were exceeded and those years when the chlorophyll *a* thresholds were not exceeded.

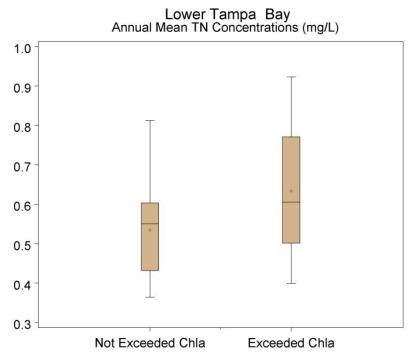


Figure 5. Comparison of TN concentrations in Lower Tampa Bay in those years when the chlorophyll *a* thresholds were exceeded and those years when the chlorophyll *a* thresholds were not exceeded.

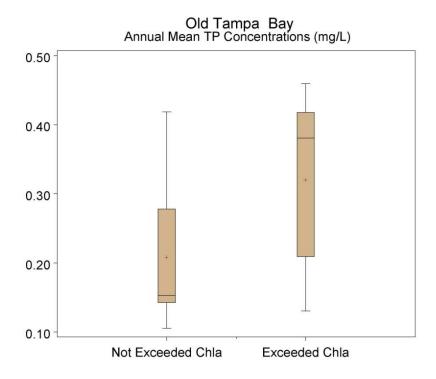


Figure 6. Comparison of TP concentrations in Old Tampa Bay in those years when the chlorophyll *a* thresholds were exceeded and those years when the chlorophyll *a* thresholds were not exceeded.

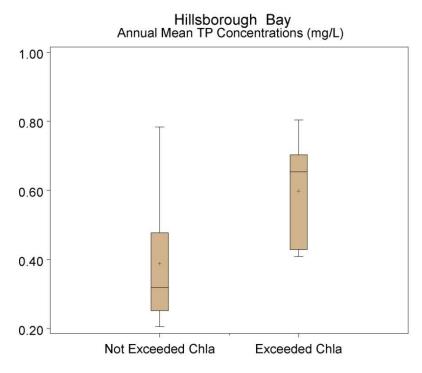


Figure 7. Comparison of TP concentrations in Hillsborough Bay in those years when the chlorophyll *a* thresholds were exceeded and those years when the chlorophyll *a* thresholds were not exceeded.

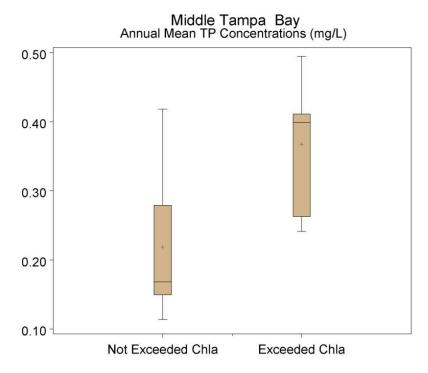


Figure 8. Comparison of TP concentrations in Middle Tampa Bay in those years when the chlorophyll *a* thresholds were exceeded and those years when the chlorophyll *a* thresholds were not exceeded.

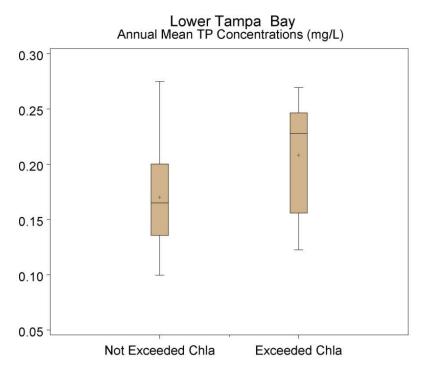


Figure 9. Comparison of TP concentrations in Lower Tampa Bay in those years when the chlorophyll *a* thresholds were exceeded and those years when the chlorophyll *a* thresholds were not exceeded.

3.3 Reference Period TN and TP Concentrations as Concentration Criteria

The analyses described above indicated that while there were significant relationships between monthly nutrient concentrations and loads, and between monthly chlorophyll a and nutrient concentrations, these relationships explained very little of the observed variation in the dependent constituent. The evaluations of annual average chlorophyll a and annual TN and TP concentrations did provide evidence that for some segments, there were differences in nutrient concentrations between those periods in which chlorophyll a targets were met and those in which they were exceeded. Based on these findings, establishment of nutrient criteria should be linked to the annual chlorophyll a and loading targets already established for Tampa Bay.

The fourth approach to developing concentration-based numeric nutrient criteria is the reference period approach. Segment-specific chlorophyll a **targets** (values at this level or below indicate desirable conditions) have been previously established (Janicki and Wade, 1996). These targets were based on a 1992-1994 reference period. In 2000, a protocol for assessing whether the Tampa Bay segments were achieving these targets was developed (Janicki and Pribble, 2000). This protocol, referred to as the Decision Matrix approach, considered the year-to-year variability in chlorophyll a concentrations and arrived at segment-specific chlorophyll a **thresholds** (values above this level indicate undesirable conditions). The threshold was the sum of the chlorophyll a target and 2X the standard error of the long-term chlorophyll a concentrations. FDEP has adopted these thresholds to assess compliance with the Tampa Bay Reasonable Assurance.

Following this approach, numeric nutrient criteria for the four bay segments can be estimated. The 1992-1994 annual geometric mean concentrations are:

•	Old Tampa Bay	TN = 0.75 mg/L	TP = 0.25 mg/L
•	Hillsborough Bay	TN = 0.82 mg/L	TP = 0.37 mg/L
•	Middle Tampa Bay	TN = 0.69 mg/L	TP = 0.24 mg/L
•	Lower Tampa Bay	TN = 0.57 mg/L	TP = 0.08 mg/L

The following are the standard deviations of the mean annual TN and TP concentrations from the 1992-2009 period (which includes all years since the chlorophyll a concentration and TN loading targets were established):

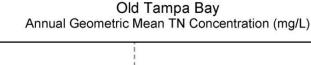
•	Old Tampa Bay	TN = 0.18 mg/L	TP = 0.06 mg/L
•	Hillsborough Bay	TN = 0.19 mg/L	TP = 0.08 mg/L
•	Middle Tampa Bay	TN = 0.18 mg/L	TP = 0.05 mg/L
•	Lower Tampa Bay	TN = 0.17 mg/L	TP = 0.02 mg/L

In the same manner as the chlorophyll *a* thresholds were developed for Tampa Bay as described above, the proposed concentration-based TN and TP criteria are defined as the sum of the concentration targets and the standard deviation of the long-term mean annual TN and TP concentrations. Table 6 presents these proposed criteria.

Table 6. Proposed numeric nutrient criteria based on geometric mean of annual concentrations for the period 1992-1994 and the standard deviation of the long-term TN and TP concentrations.

Segment	TN Concentration (mg/L)	TP Concentration (mg/L)
Old Tampa Bay	0.93	0.31
Hillsborough Bay	1.01	0.45
Middle Tampa Bay	0.87	0.29
Lower Tampa Bay	0.74	0.10

The proposed TN and TP concentration criteria are compared to the observed geometric mean annual TN and TP concentrations in Figures 10 through 17. The horizontal lines represent the proposed criteria; the data to the right of the vertical lines depict the TN and TP concentrations since the establishment of the reference period (1992-1994).



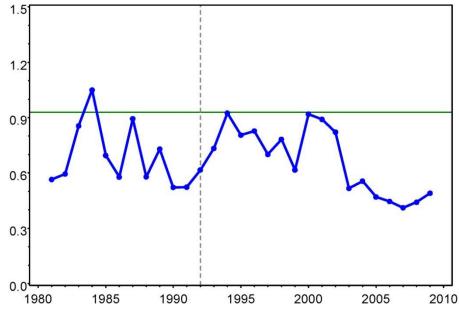


Figure 10. Comparison of proposed TN concentration criterion for Old Tampa Bay to the annual geometric mean TN concentrations from 1981 through 2009.

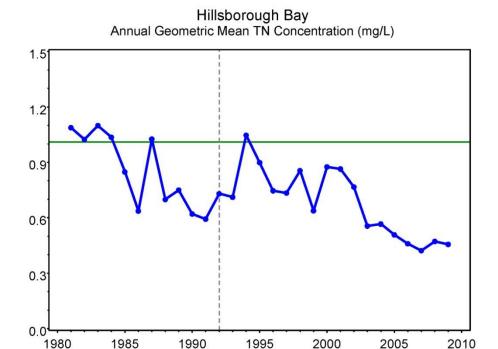


Figure 11. Comparison of proposed TN concentration criterion for Hillsborough Bay to the annual geometric mean TN concentrations from 1981 through 2009.

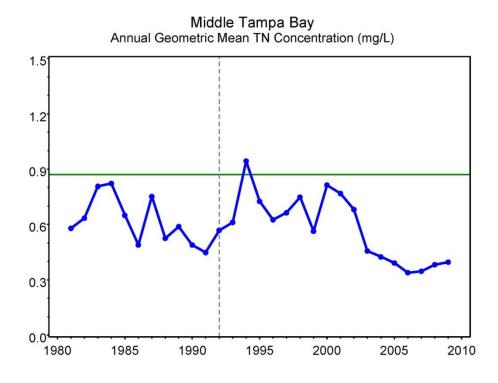


Figure 12. Comparison of proposed TN concentration criterion for Middle Tampa Bay to the annual geometric mean TN concentrations from 1980 through 2009.

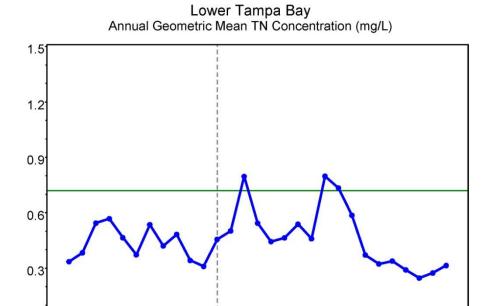


Figure 13. Comparison of proposed TN concentration criterion for Lower Tampa Bay to the annual geometric mean TN concentrations from 1980 through 2009.

0.0

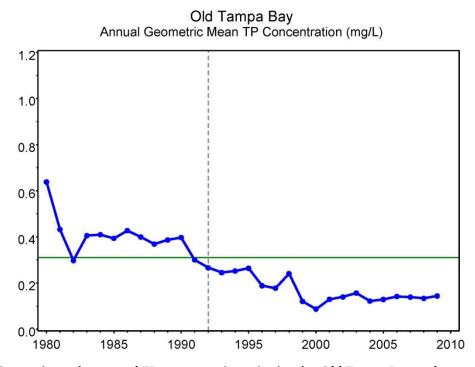


Figure 14. Comparison of proposed TP concentration criterion for Old Tampa Bay to the annual geometric mean TP concentrations from 1980 through 2009.

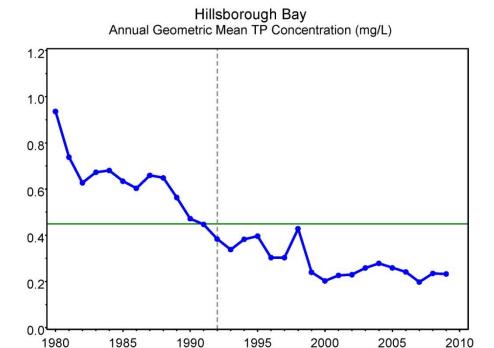


Figure 15. Comparison of proposed TP concentration criterion for Hillsborough Bay to the annual geometric mean TP concentrations from 1980 through 2009.

1995

2000

2005

2010

1985

1980

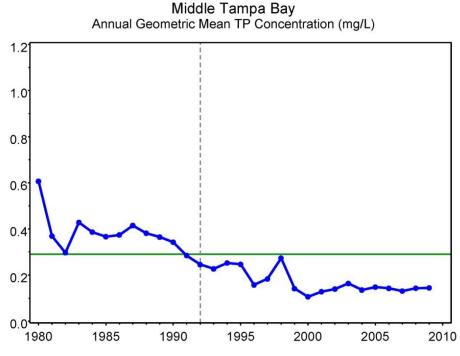
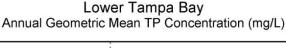


Figure 16. Comparison of proposed TP concentration criterion for Middle Tampa Bay to the annual geometric mean TP concentrations from 1980 through 2009.

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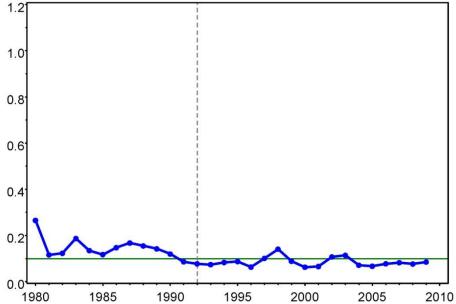


Figure 17. Comparison of proposed TP concentration criterion for Lower Tampa Bay to the annual geometric mean TP concentrations from 1980 through 2009.

4.0 Conclusions

The following conclusions can be drawn from the analyses and results discussed above:

- Tampa Bay is nitrogen-limited as indicated by both ambient TN:TP ratios and nutrient addition bioassays.
- There is no discernable relationship between TN loadings and in-bay TN concentrations or between TP loadings and in-bay TP concentrations in any bay segment. This is not because loadings do not affect in-bay concentrations, but because various other confounding factors, for which sufficient data are not available, play a role in relationships between loadings and concentrations
- On a monthly time scale, the relationships between either TN concentrations or TP
 concentrations and chlorophyll a concentrations do not explain a significant proportion of
 the variability in the chlorophyll a concentrations to support development of concentrationbased numeric nutrient criteria in any bay segment.
- There are differences in TN concentrations and TP concentrations, particularly in Hillsborough Bay and Middle Tampa Bay, in those years when the chlorophyll a concentration thresholds are met when compared to those observed in years when the chlorophyll a concentration thresholds are not met. However, there is a great deal of variability within the data obtained in either group of years and these differences are not

recommended as the basis for the establishment of concentration-based numeric nutrient criteria for Tampa Bay.

• The reference period approach is recommended for the establishment of concentration-based TN and TP criteria for Tampa Bay. The segment-specific annual geometric mean nitrogen and phosphorus concentrations of the 1992-1994 period were increased by one standard deviation (as derived from 1992-2009 data) to develop the proposed concentration numeric nutrient criteria. These criteria are:

-	Old Tampa Bay	TN = 0.93 mg/L	TP = 0.31 mg/L
-	Hillsborough Bay	TN = 1.01 mg/L	TP = 0.45 mg/L
-	Middle Tampa Bay	TN = 0.87 mg/L	TP = 0.29 mg/L
-	Lower Tampa Bay	TN = 0.74 mg/L	TP = 0.10 mg/L.

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Attachment 1 Results of City of Tampa Bay Study Group Bioassays (Johansson, 2009)

NUTRIENT ENRICHMENT STUDIES OF NATURAL PHYTOPLANKTON POPULATIONS IN TAMPA BAY

A SUMMARY OF RESULTS JUNE 1993 TO AUGUST 2009

J.O.R. JOHANSSON THE CITY OF TAMPA WASTEWATER DEPARTMENT BAY STUDY GROUP

NOVEMBER 16, 2009

NUTRIENT ENRICHMENT STUDIES OF NATURAL PHYTOPLANKTON POPULATIONS IN TAMPA BAY. A SUMMARY OF RESULTS JUNE 1993 TO AUGUST 2009

INTRODUCTION

The Bay Study Group (BSG) has been conducting nutrient enrichment studies (bioassays) on natural phytoplankton populations at four locations in Tampa Bay since 1993 (Figure 1). This report will summarize findings for all tests that have been performed to date, from June 1993 through August 2009.

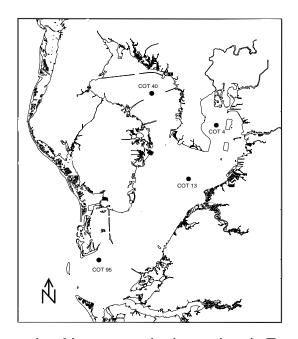


Figure 1. Phytoplankton nutrient bioassay monitoring stations in Tampa Bay.

METHODS

The bioassays have been performed on the natural phytoplankton population collected from surface waters of the four stations, with one station located in each of the four major segment of Tampa Bay, as shown below:

- Hillsborough Bay COT4
- Old Tampa Bay COT40
- Middle Tampa Bay COT13
- Lower Tampa Bay COT95

Bioassay measurements were performed on a quarterly schedule from June 1993 through August 1995 for all stations except COT95. At this station monthly tests were conducted during the first year of the program. From February 1996 to the present, bioassays have been performed twice per year, during late winter and late summer.

The bioassay method used is similar to a method used in Chesapeake Bay waters (see Fisher et al. 1992a and b). An outline of the specific method used by the BSG is provided here.

A large volume of surface water is collected for the following nutrient treatments. Each treatment is conducted in duplicate on 3l samples:

- Controls (no nutrient addition).
- N-additions (NH3-N added to reach a final concentration in the sample of near 50uM).
- P-additions (PO4-P added to reach a final concentration in the sample of near 5uM).
- N+P-additions (combination of the N-additions and P-additions).

The treatment samples are incubated under natural sunlight in ambient bay water temperatures for an appropriate incubated period (usually 48h for the late winter and 24h for the late summer tests).

The growth response of the natural phytoplankton community to the different treatments is determined through measurements in changes of algal biomass, measured as chlorophyll-a.

Paired t-test statistics (p<0.05) and non-statistical evaluations of treatment responses are used to interpret the bioassay results and to group the growth response to the nutrient additions into the following response categories:

- Exclusive N limitation: (1) the addition of P induced no response relative the control, and (2) the addition of N alone had virtually the same effect as the addition of N+P.
- Primary N limitation: (1) the addition of P alone induced little response relative the control, (2) the addition of N alone induced a response, and (3) the addition of N+P induced the largest response.
- Balanced NP limitation: (1) the addition of N and P alone induced no response relative the control, (2) the addition of N+P induced a large response.
- Exclusive P limitation: (1) the addition of N induced no response relative the control, and (2) the addition of P alone had virtually the same effect as the addition of N+P.
- Primary P limitation: (1) the addition of N alone induced little response relative the control, (2) the addition of P alone induced a response, and (3) the addition of N+P induced the largest response.
- No response to any nutrient addition, indicating nutrient saturation, light limitation, and/or insufficient incubation time.

RESULTS

A total of 152 bioassay experiments have been conducted since the start of the program in June 1993.

The growth response of the natural phytoplankton community to the different nutrient treatments were grouped into the response categories described above. The result of these analyses is summarized in Table 1.

Table 1. Results from natural phytoplankton nutrient bioassays in the four major subsections of Tampa Bay, 1993 – 2009.

Bioassay response	Hillsborough Bay	Old Tampa Bay	Middle Tampa Bay	Lower Tampa Bay
	COT4	COT40	COT13	COT95
Exclusive N limitation	29	25	28	39
Primary N limitation	2	2	2	1
Balanced	0	0	0	0
Exclusive P limitation	0	0	0	0
Primary P limitation	0	0	0	0
No response	0	3	0	0

DISCUSSION AND CONCLUSION

One-hundred-fifty-two natural phytoplankton community nutrient limitation experiments have been conducted in Tampa Bay by the BSG from 1993 to the present. The strong nitrogen dependence by the Tampa Bay phytoplankton community is obvious in all four bay segments; of the 152 bioassay tests conducted to date, 149 indicate that nitrogen was the stronger limiting nutrient. None of the 152 tests have indicated that phosphorous was the stronger limiting nutrient. However, three tests in OTB have shown a lack of phytoplankton growth response by either N or P additions.

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Attachment 2 Data Sources

The following data sources were used in the analyses performed for this task:

- The water quality data were obtained from the Environmental Protection Commission of Hillsborough County (EPCHC). Their sampling program collects surface samples on a monthly basis from the following series of fixed stations. Among others, the primary analytes included:
 - Chlorophyll a
 - TN
 - TP
 - Salinity.

Old Tampa Bay	Hillsborough Bay	Middle Tampa Bay	Lower Tampa Bay
46	44	11	25
60	70	9	23
64	52	81	21
47	6	84	91
65	71	33	90
63	7	32	24
66	55	13	95
40	08	14	92
41	73	82	93
67	80	28	
50		16	
38		19	
51			
68			
36			

• The TN and TP loading estimates were obtained from a series of reports produced for the TBEP (1985-2002) and FDEP (2003-2007). These are:

Zarbock, H., A. Janicki, D. Wade, D. Heimbuch, and H. Wilson. 1994. Estimates of Total Nitrogen, Total Phosphorus, and Total Suspended Solids Loadings to Tampa Bay, Florida. Technical Publication #04-94 of the Tampa Bay National Estuary Program. Prepared by Coastal Environmental, Inc. Prepared for Tampa Bay National Estuary Program. St. Petersburg, FL.

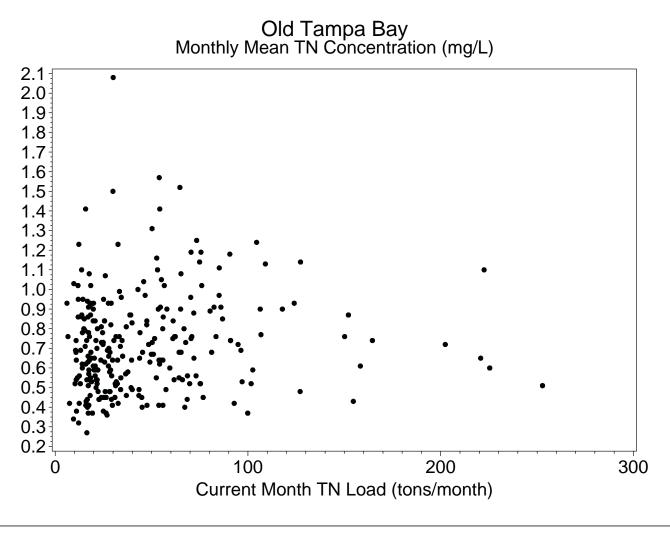
Zarbock, H.W., A.J. Janicki, and S.S. Janicki. 1996. Estimates of Total Nitrogen, Total Phosphorus, and Total Suspended Solids Loadings to Tampa, Bay, Florida. Technical Appendix: 1992-94 Total Nitrogen Loadings to Tampa Bay. Technical Publication #19-96 of the Tampa Bay National Estuary Program. Prepared by Coastal Environmental, Inc. Prepared for Tampa Bay National Estuary Program. St. Petersburg, FL.

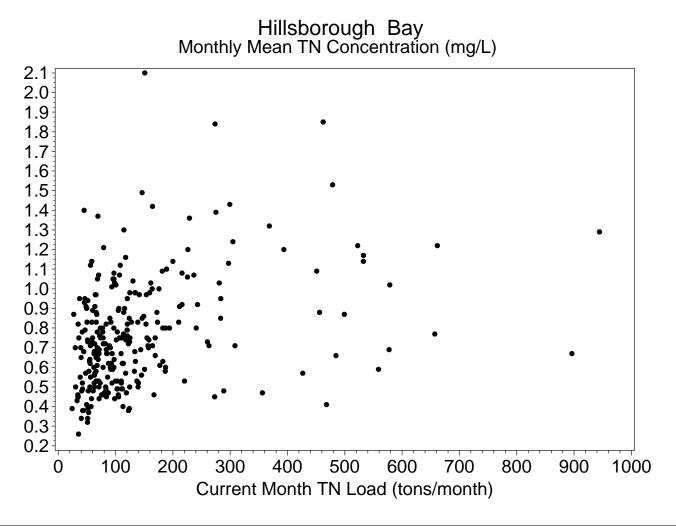
Pribble, R., A. Janicki, H. Zarbock, S. Janicki, and M. Winowitch. 2001. Estimates of Total Nitrogen, Total Phosphorus, Total Suspended Solids, and Biochemical Oxygen Demand Loadings to Tampa, Bay, Florida: 1995-1998. Technical report #05-01 of the Tampa Bay Estuary Program. Prepared by Janicki Environmental, Inc. Prepared for Tampa Bay Estuary Program. St. Petersburg, FL.

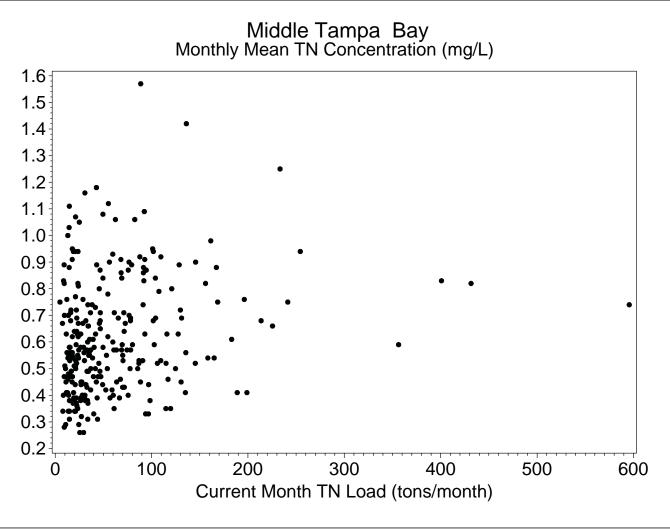
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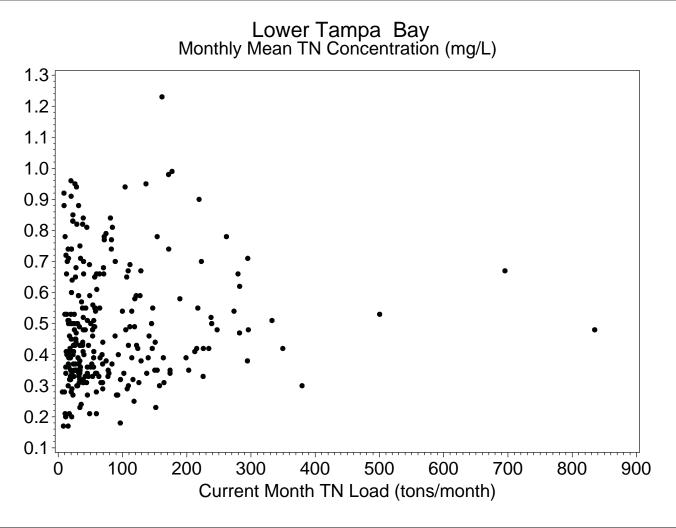
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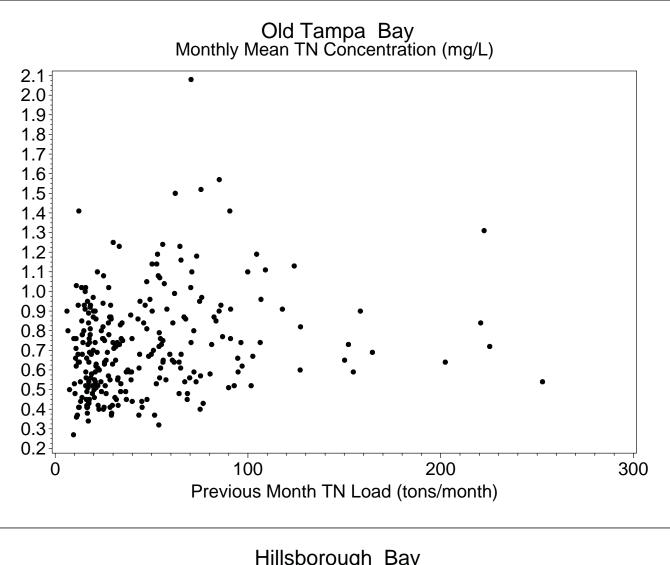
Attachment 3 Monthly TN Concentrations and Monthly TN Loads

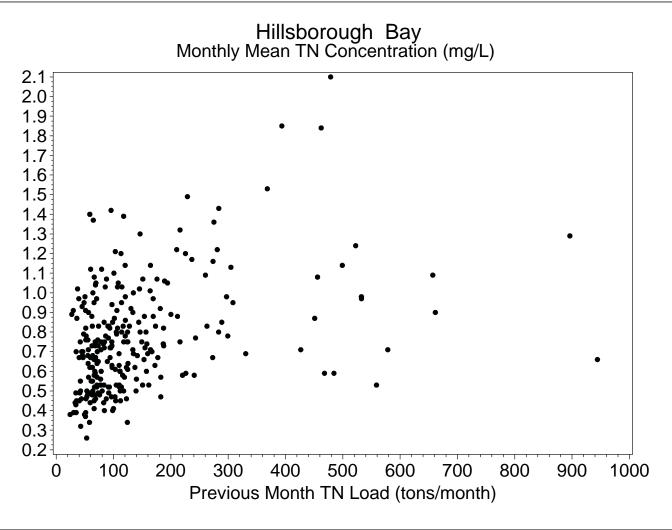


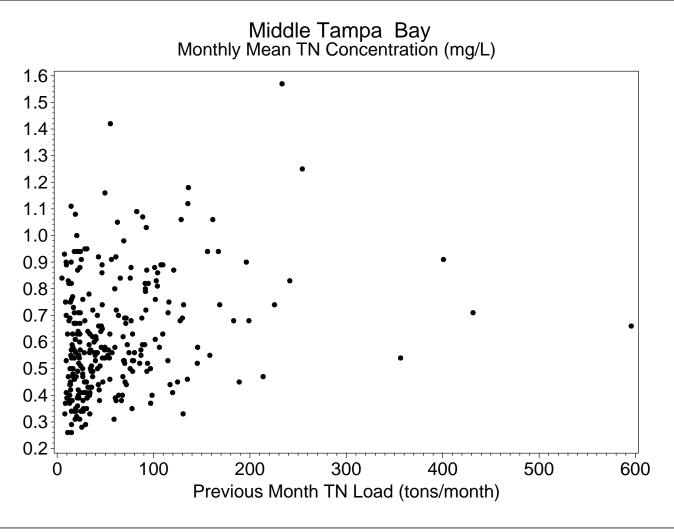


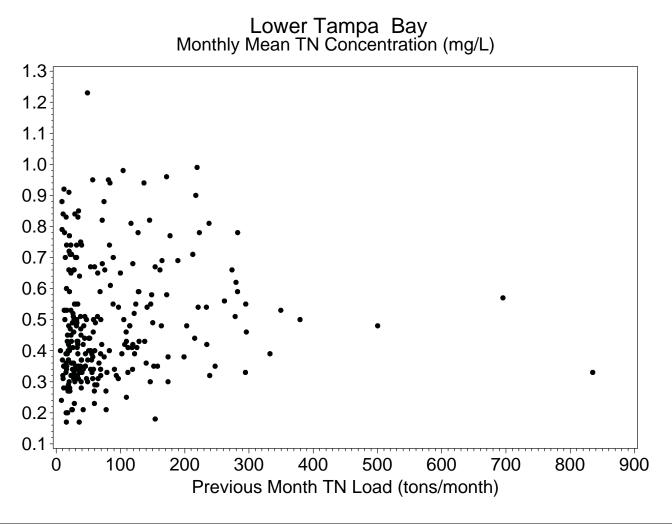


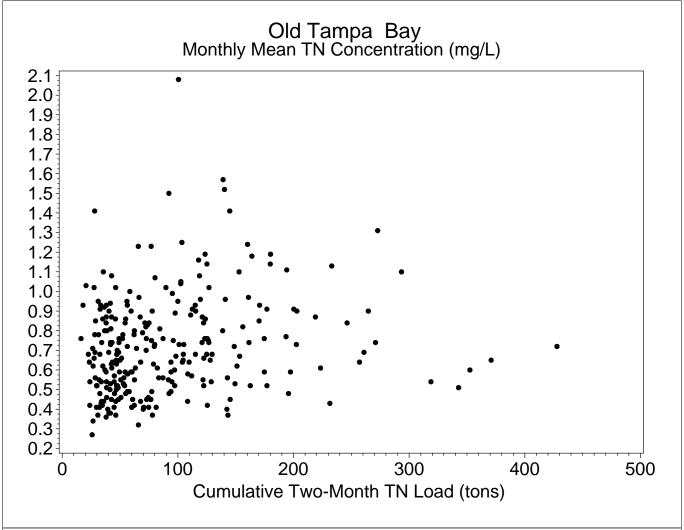


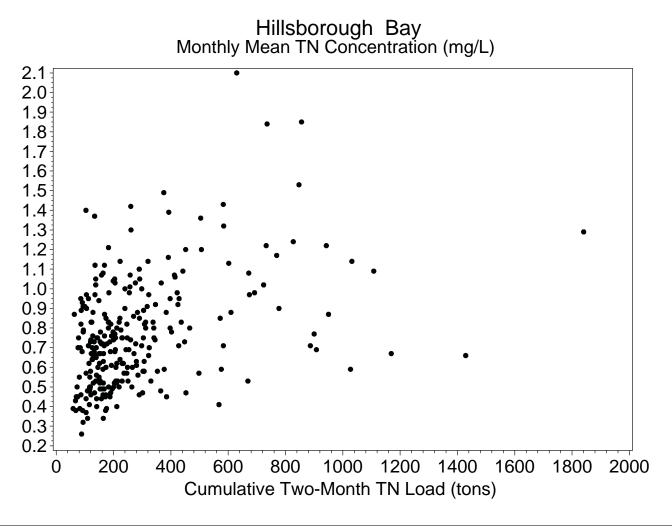


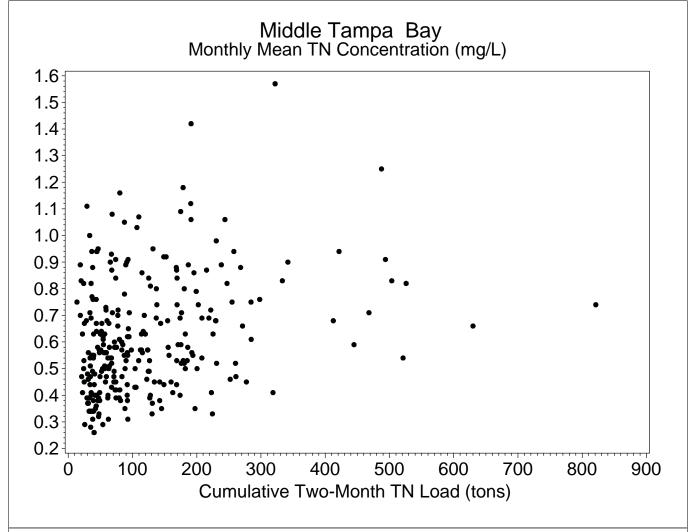


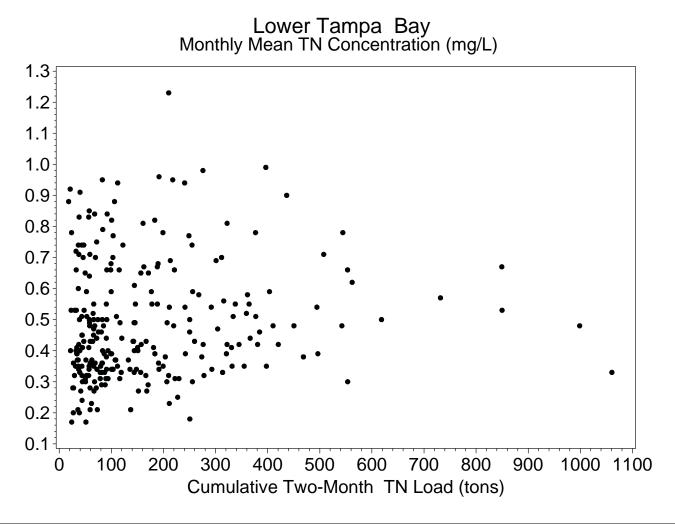


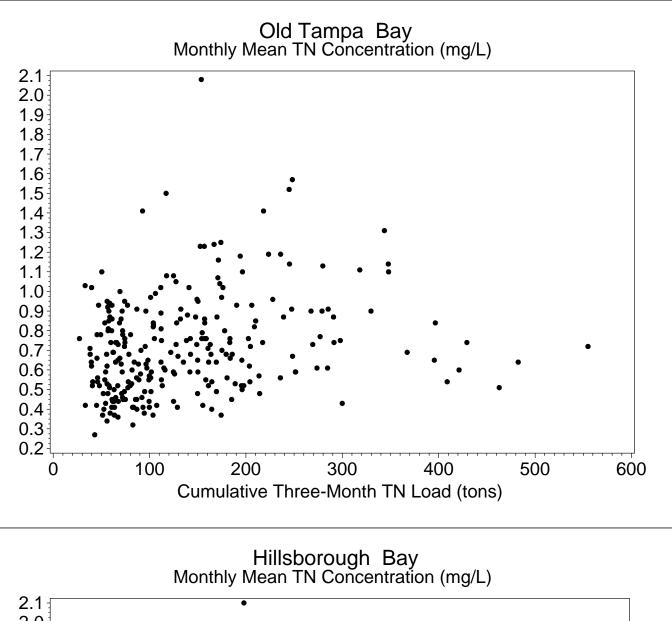


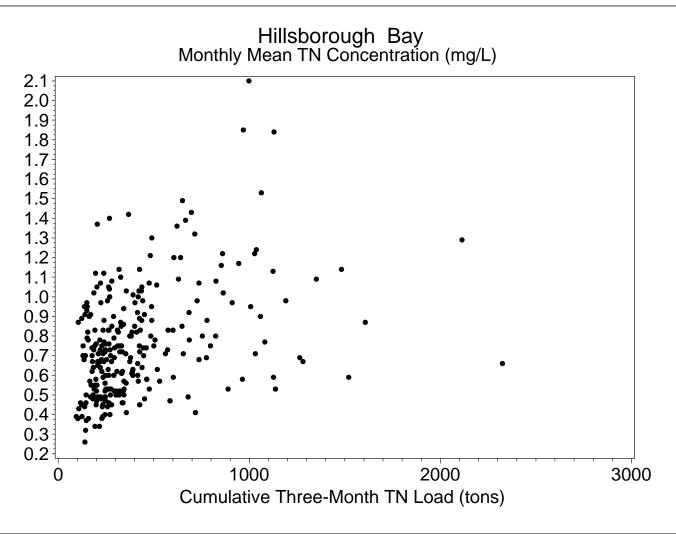


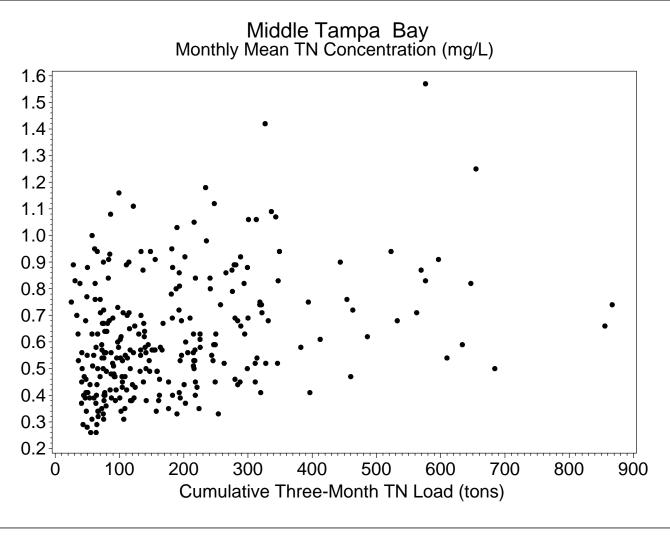


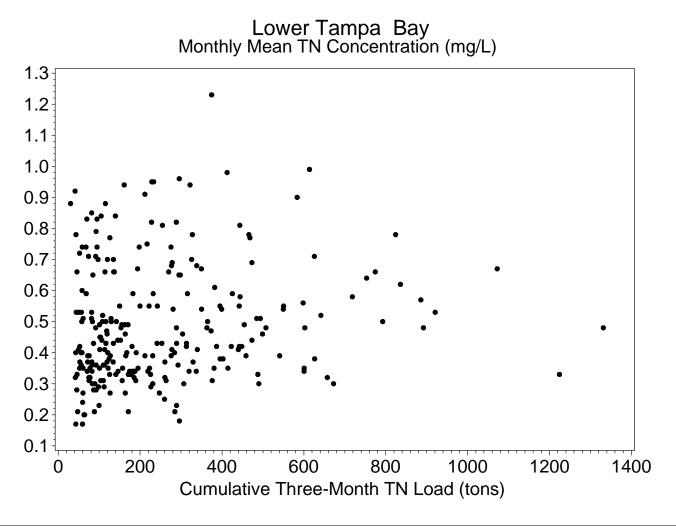


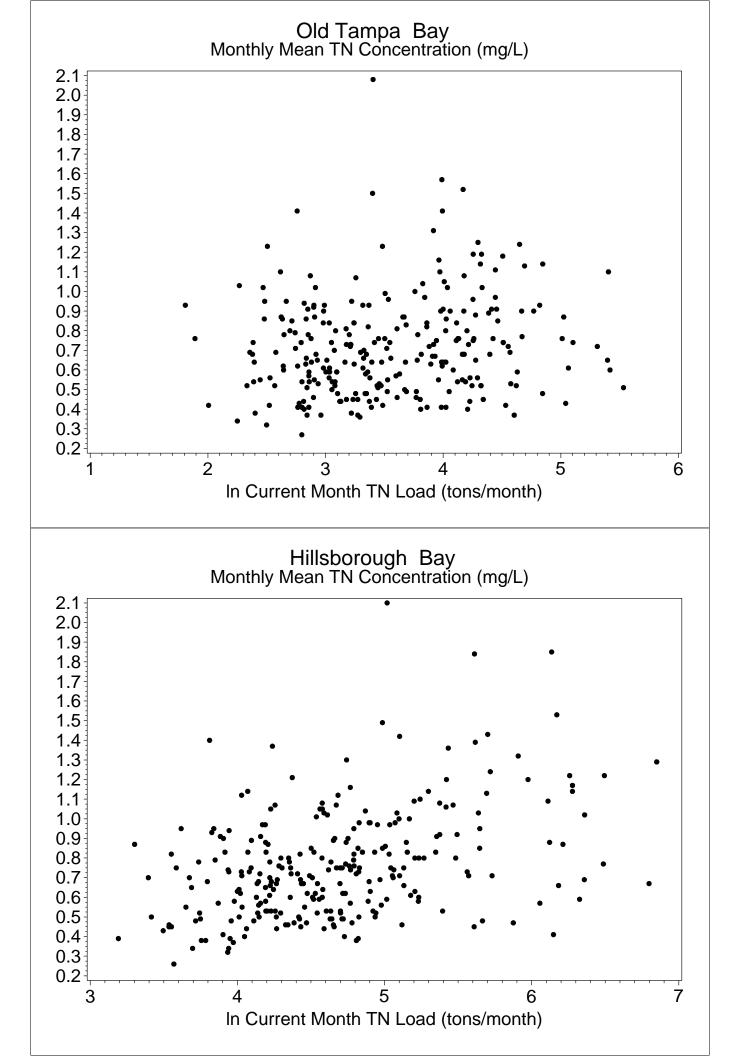


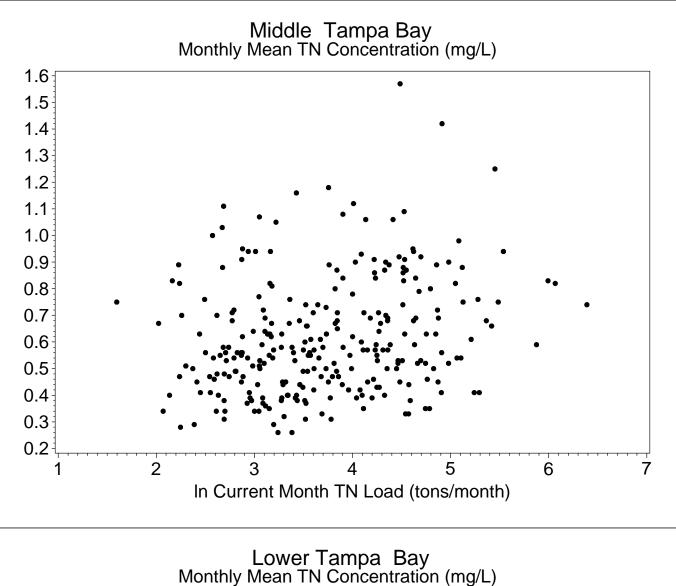


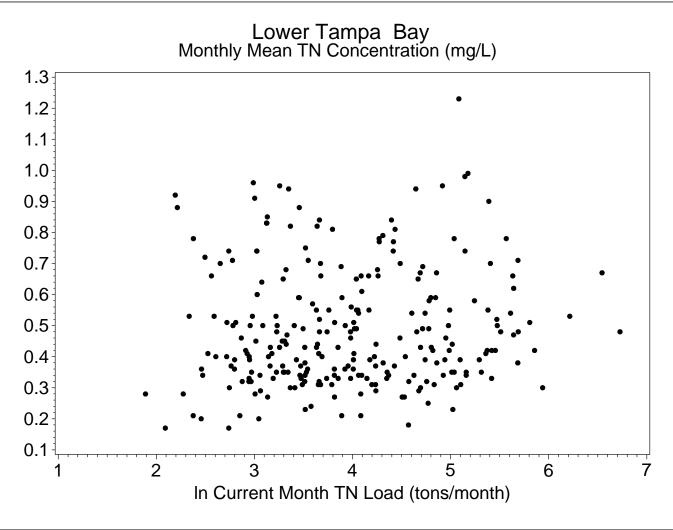


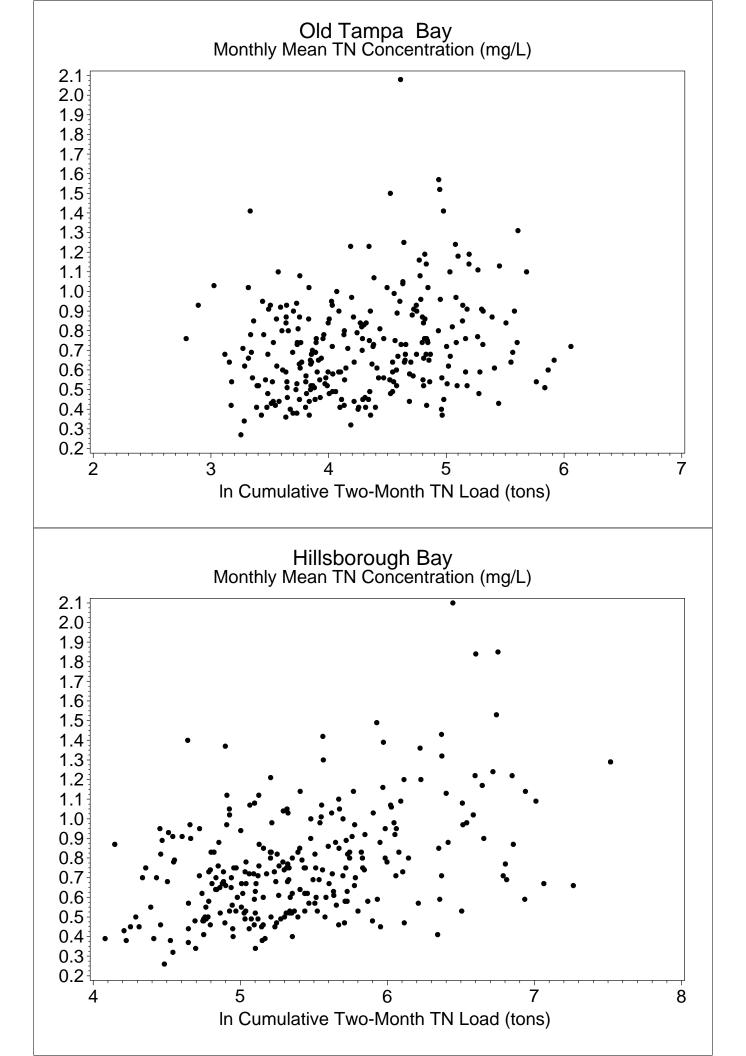


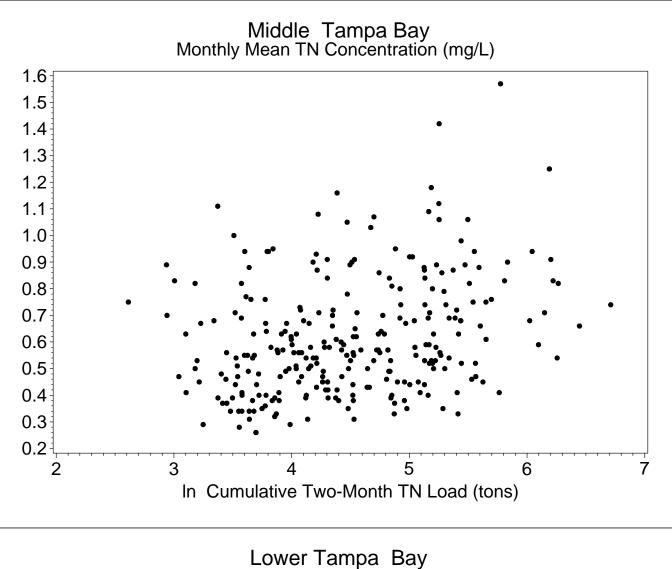


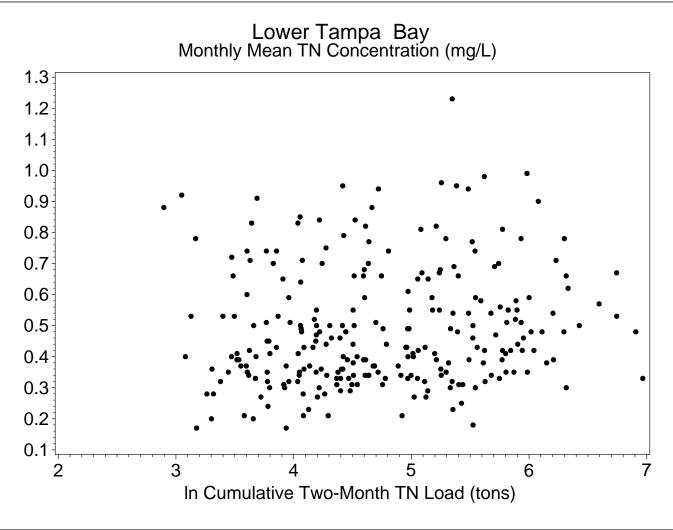


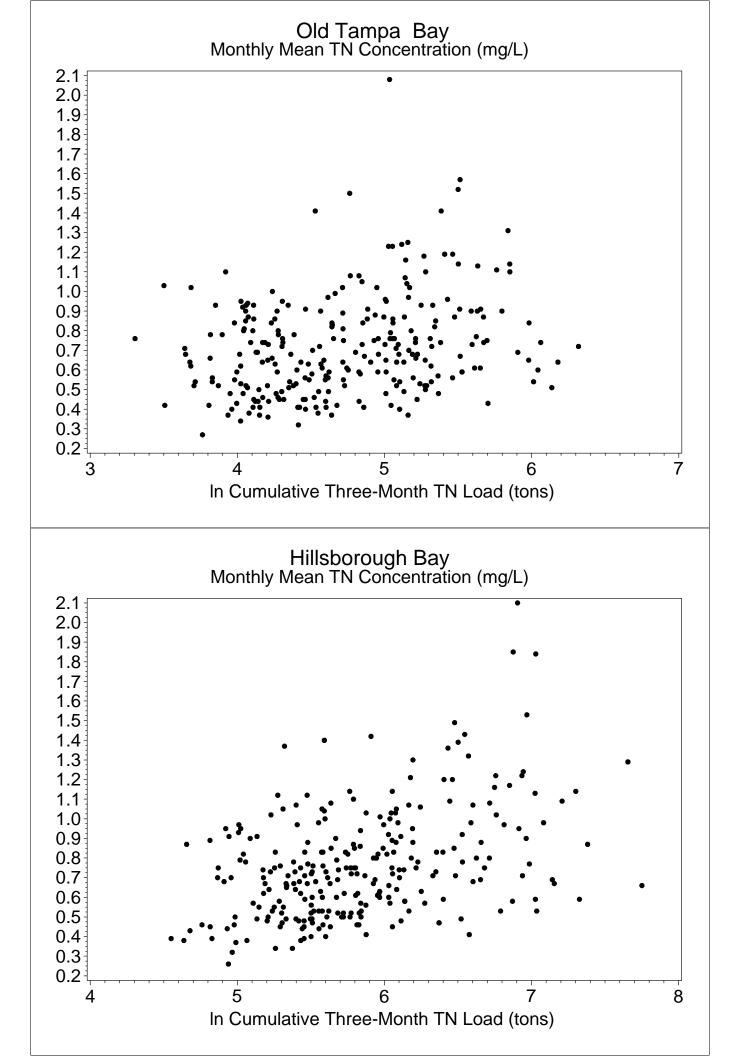


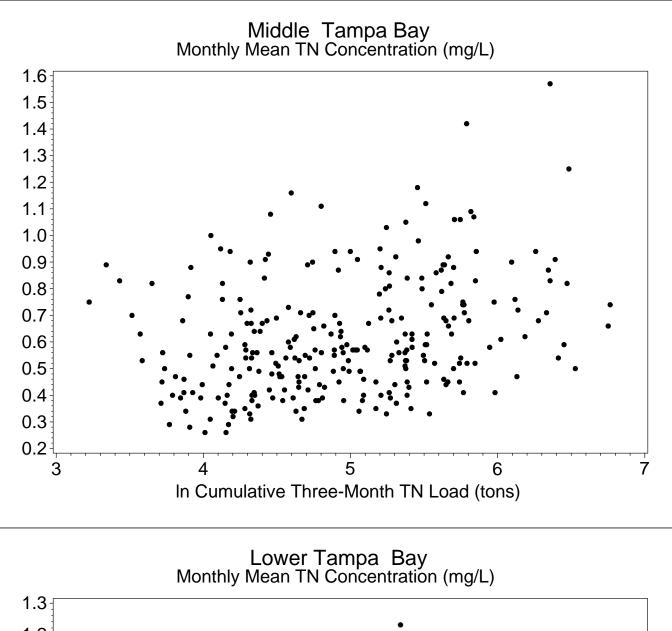


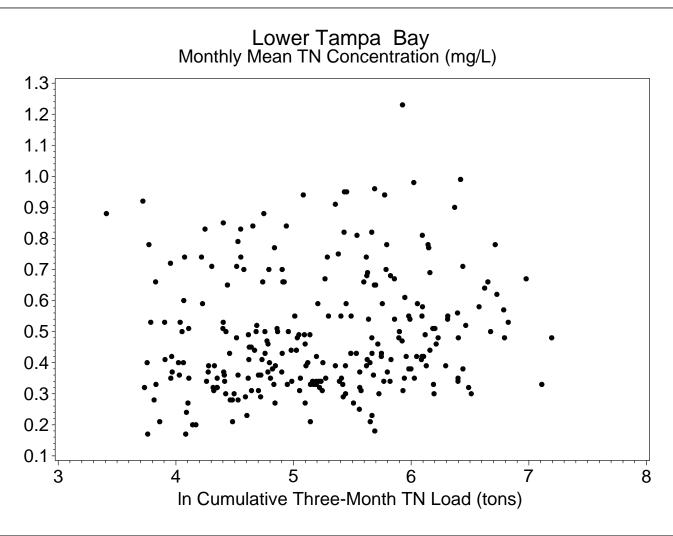




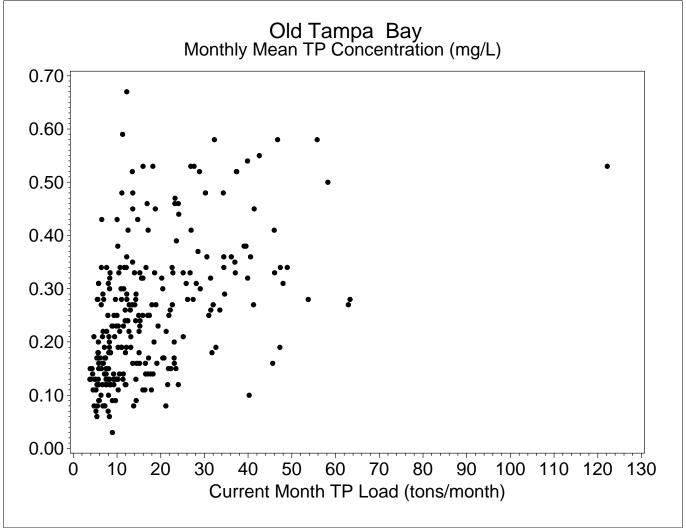


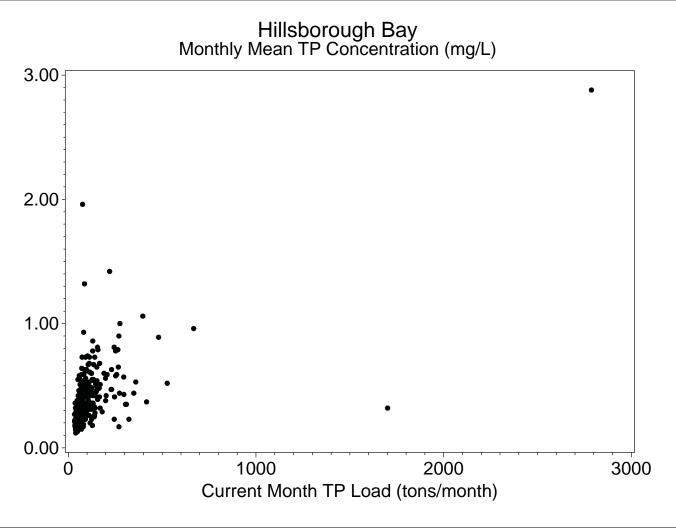


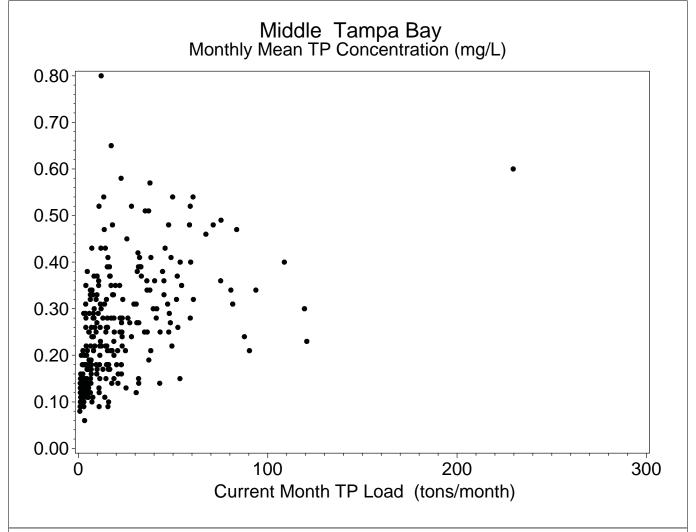


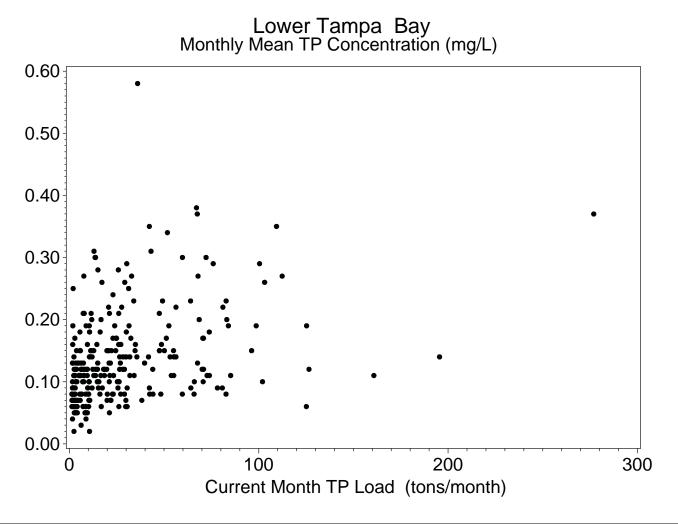


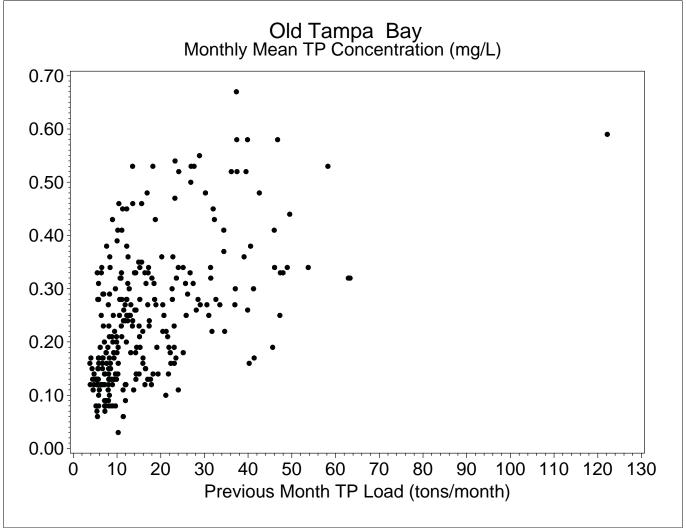
Attachment 4 Monthly TP Concentrations and Monthly TP Loads

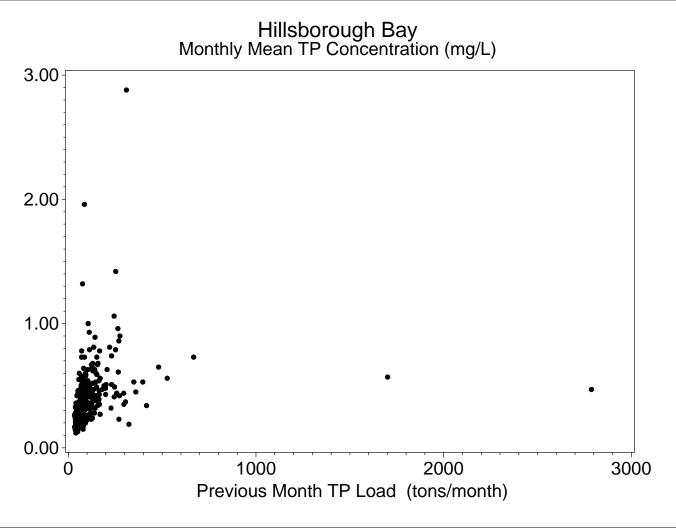


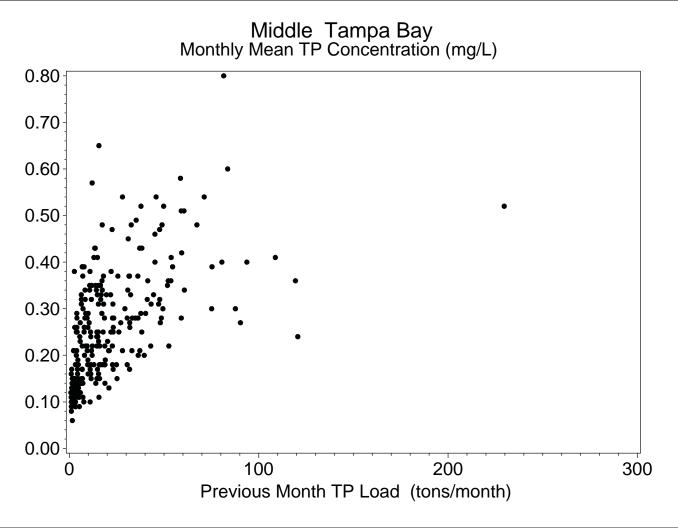


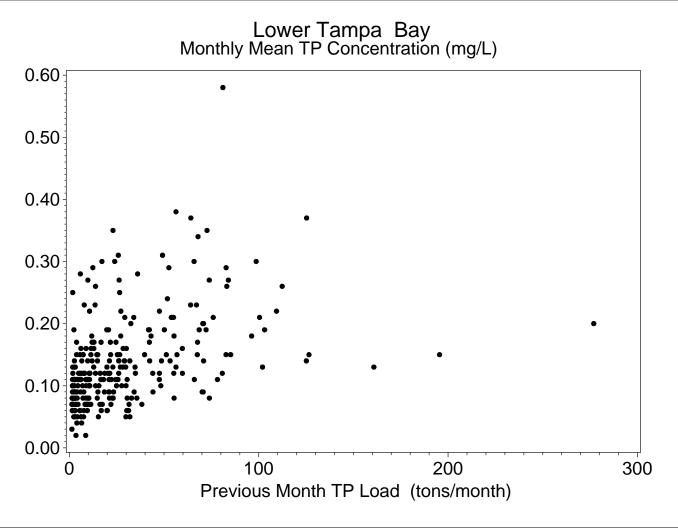


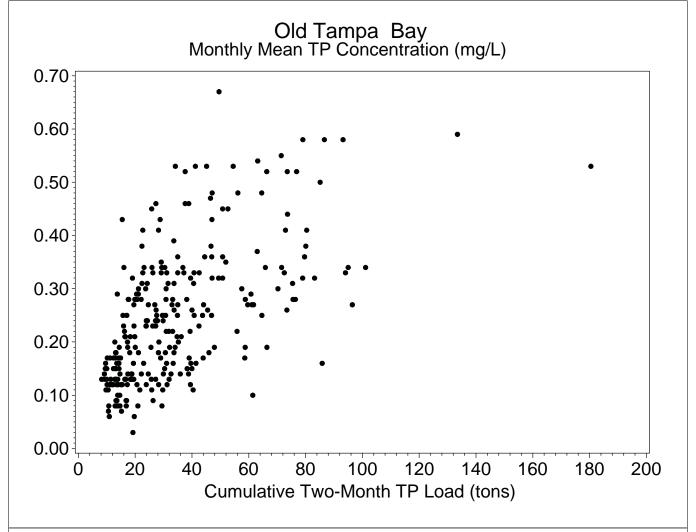


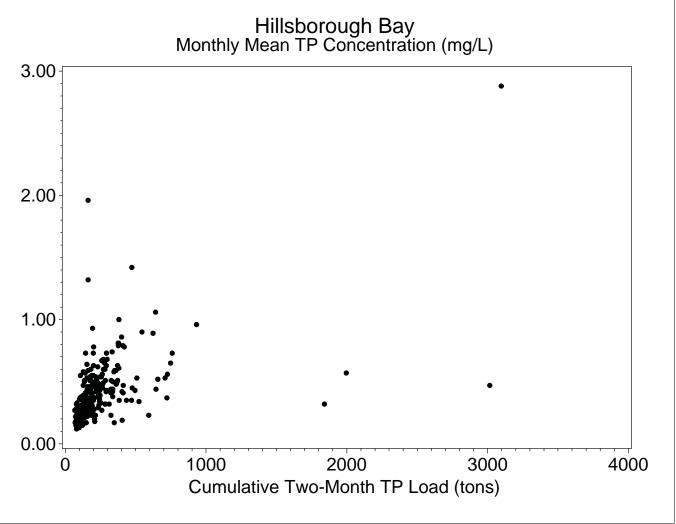


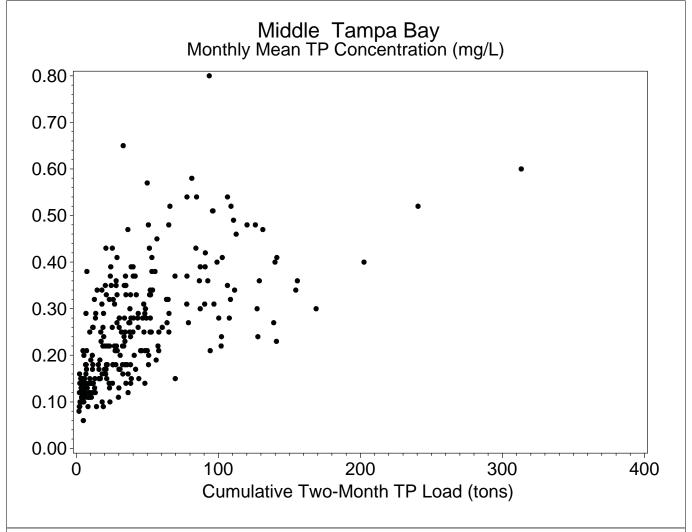


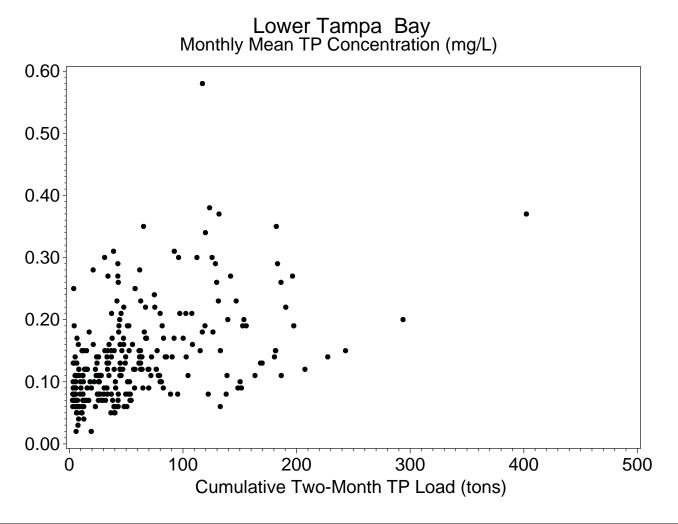


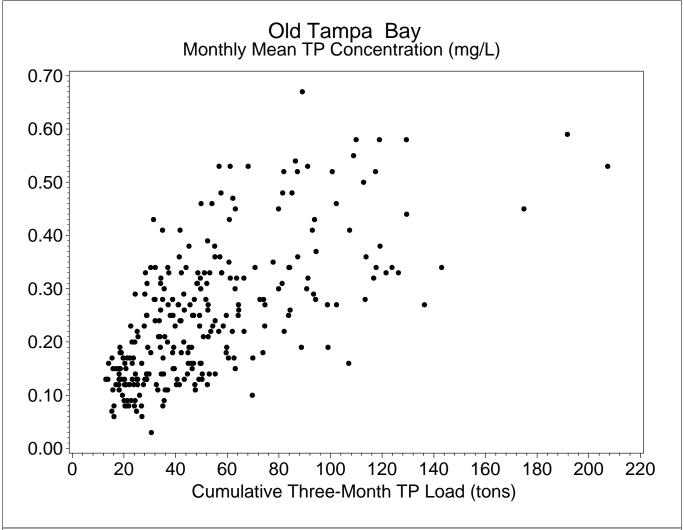


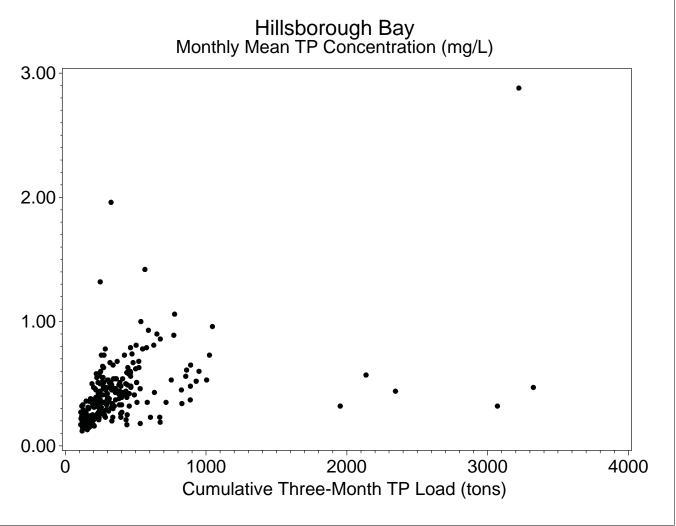


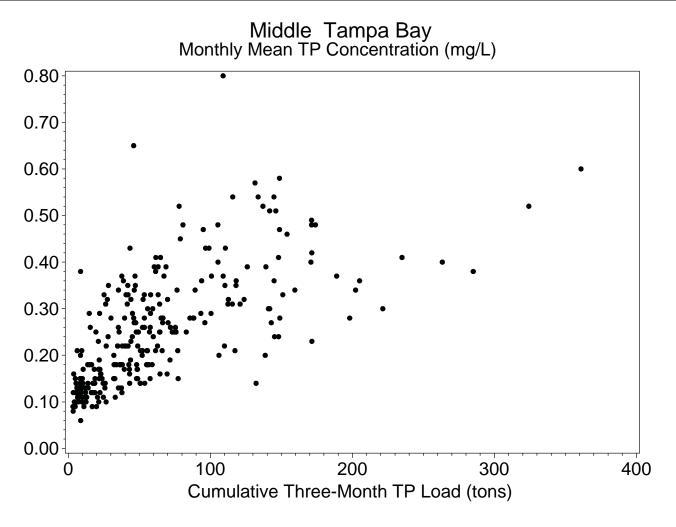


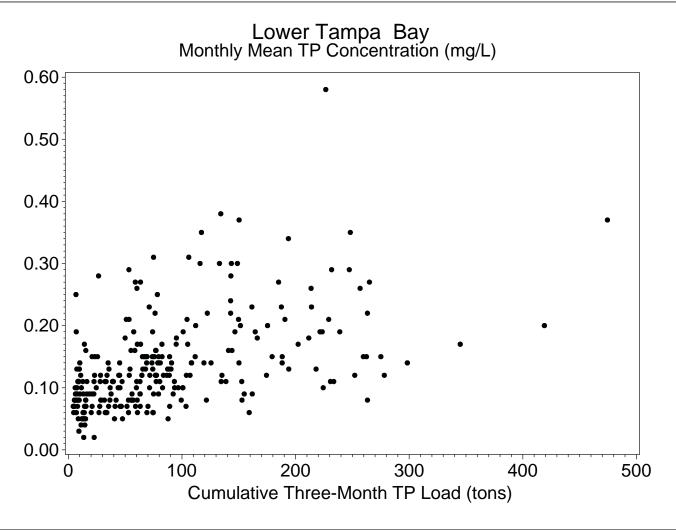


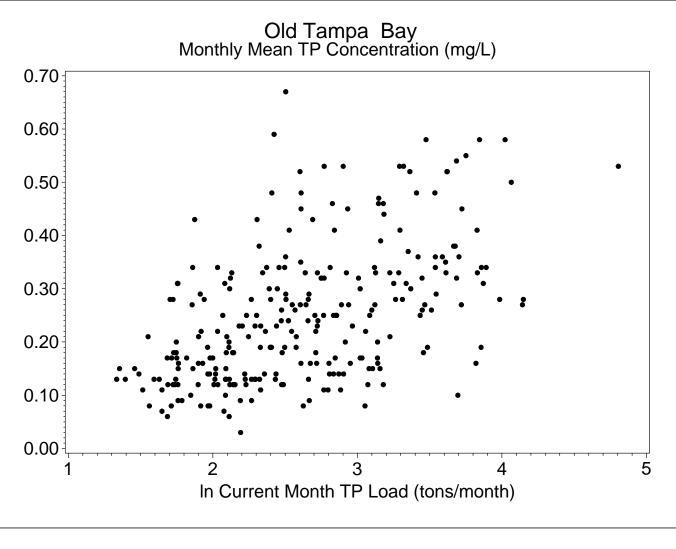


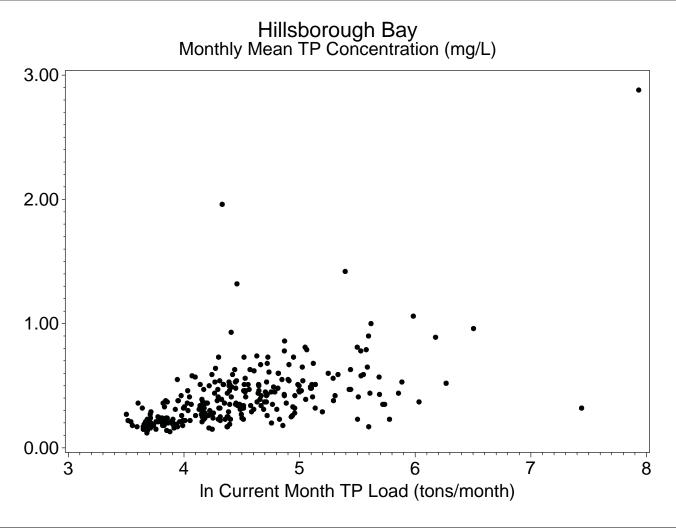


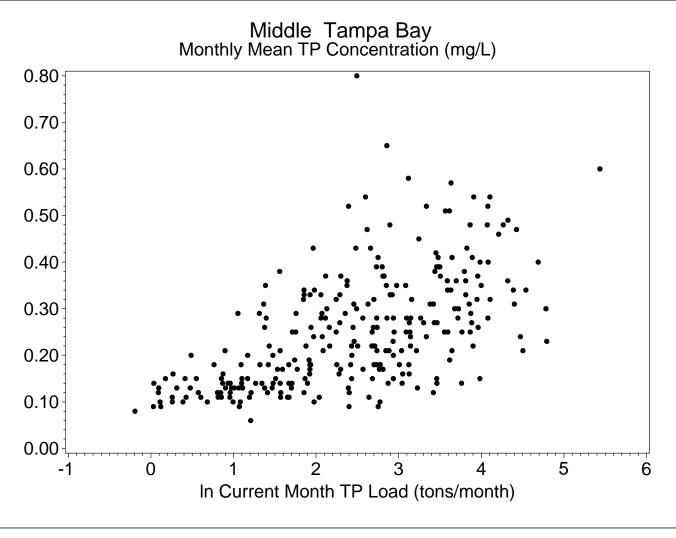


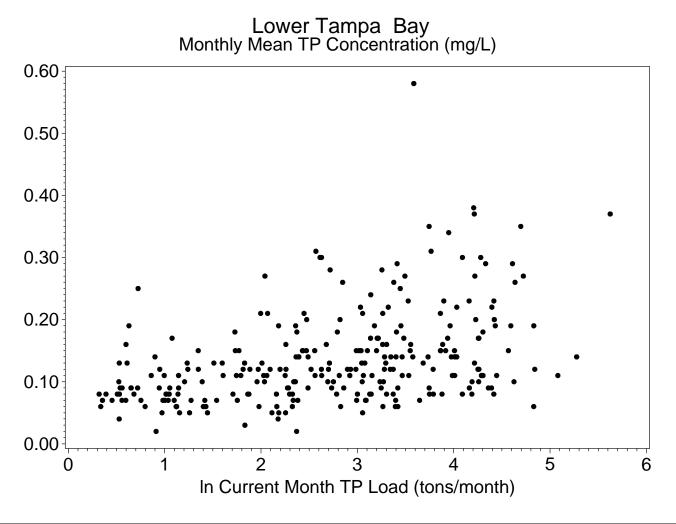


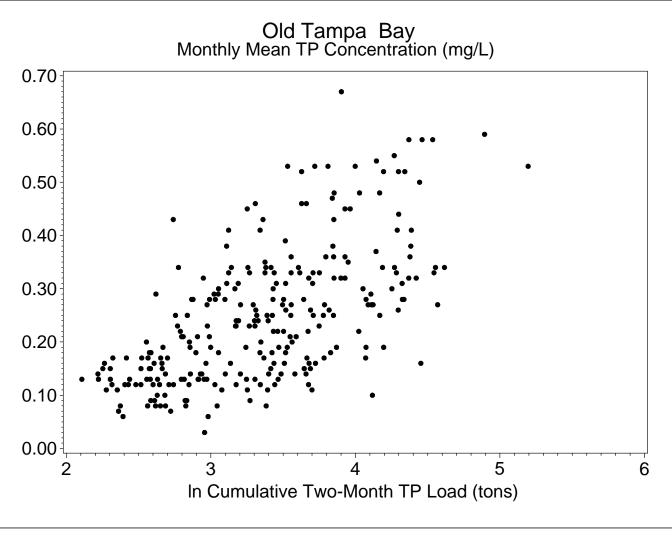


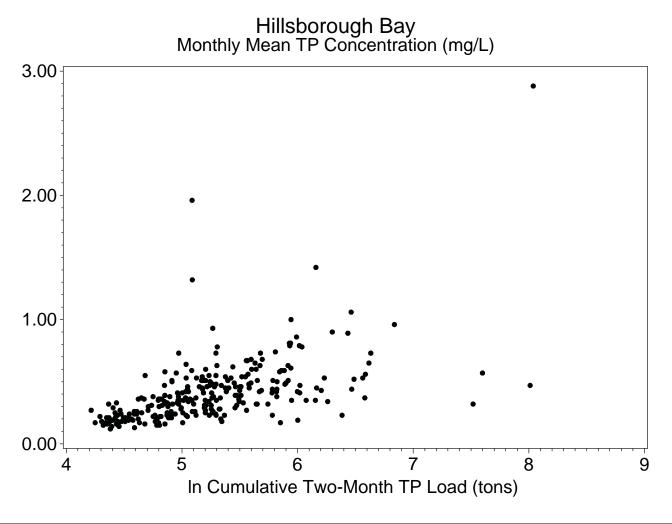


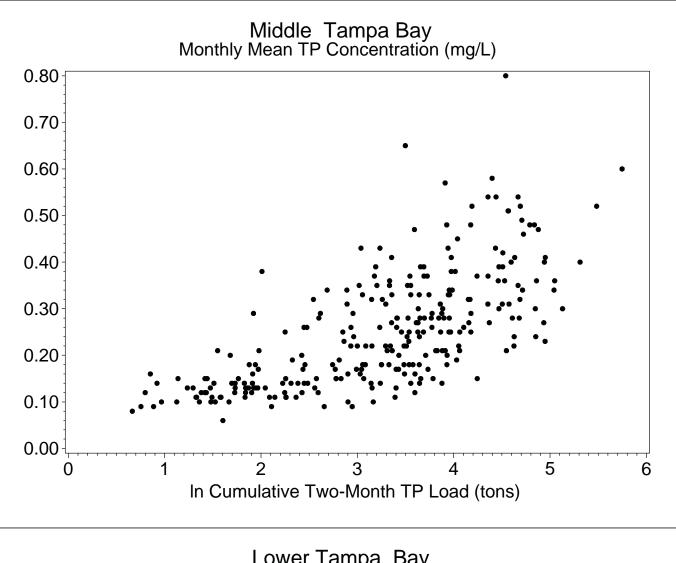


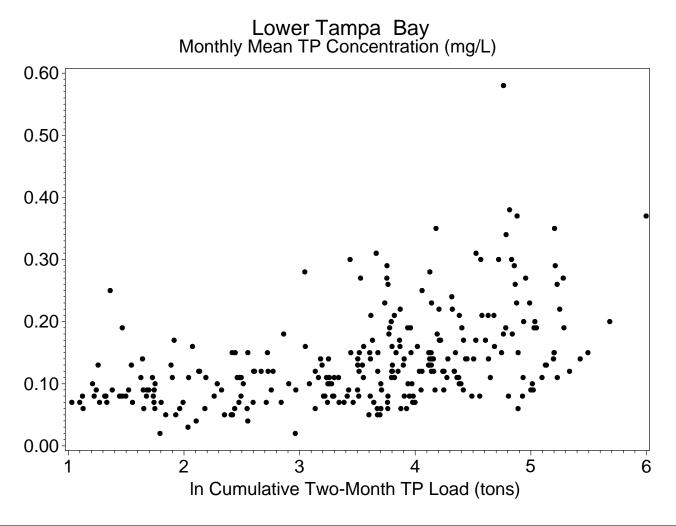


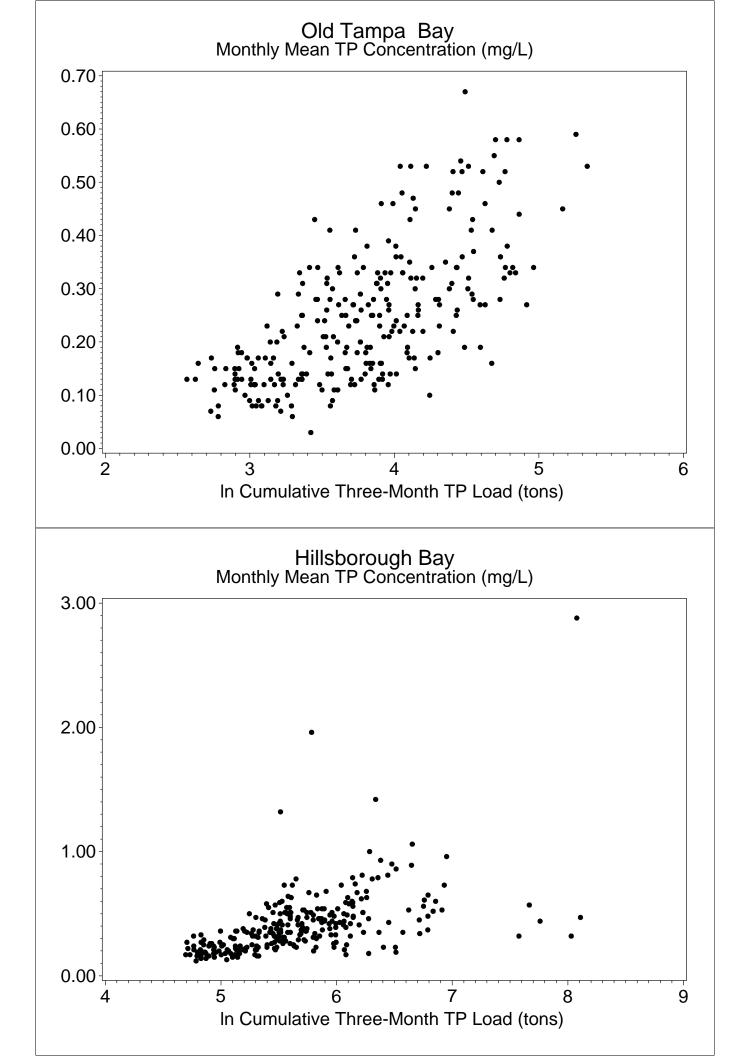


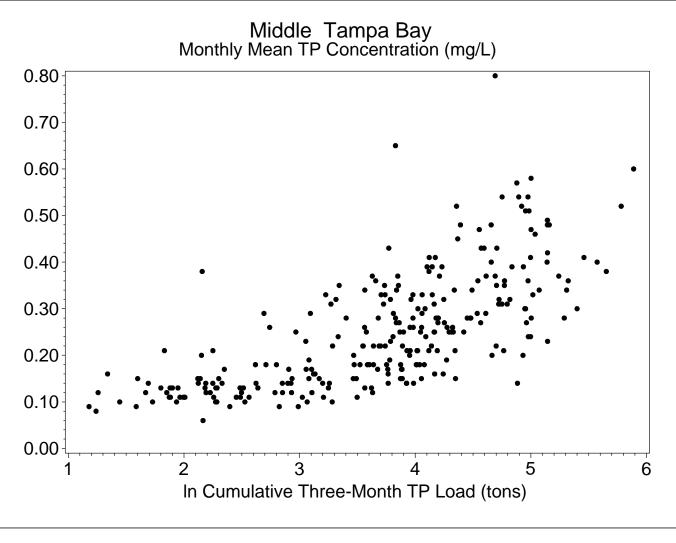


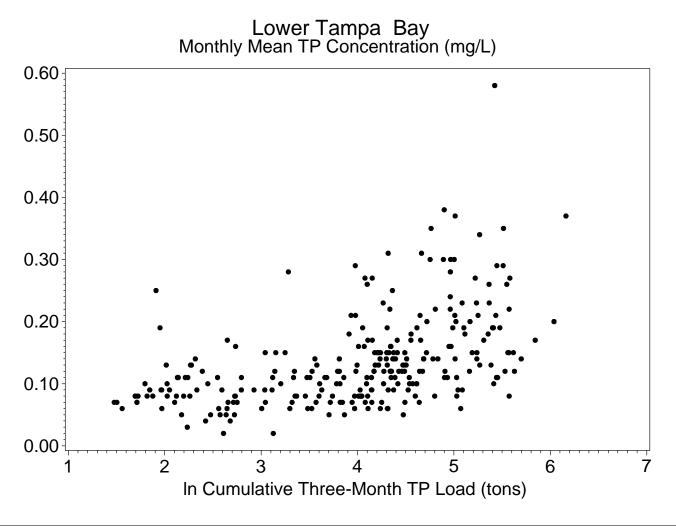




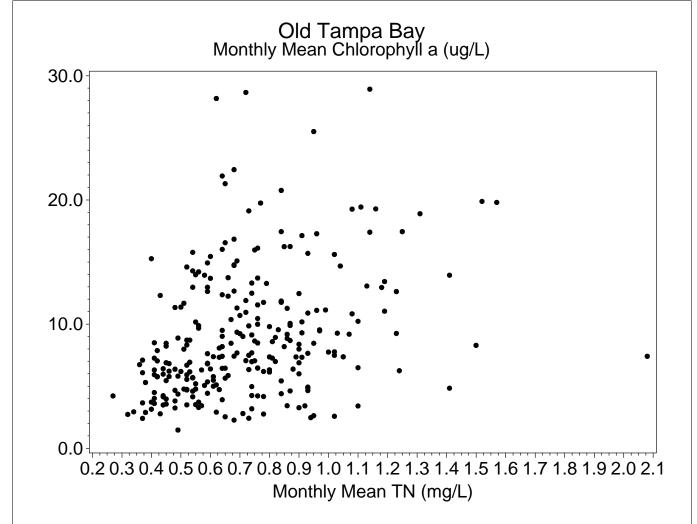


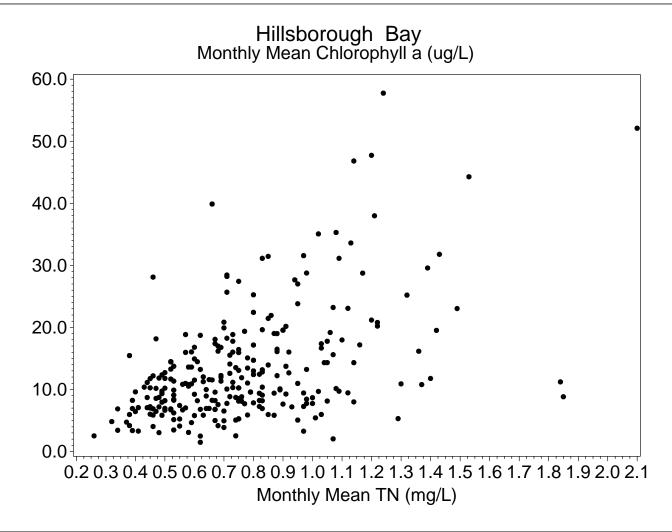


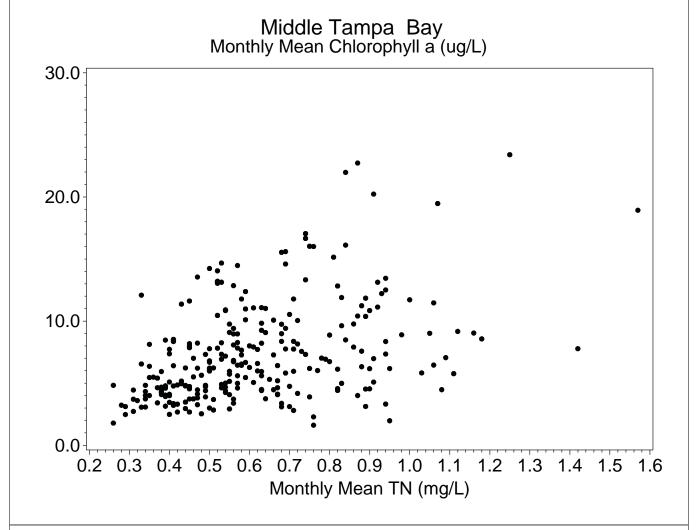


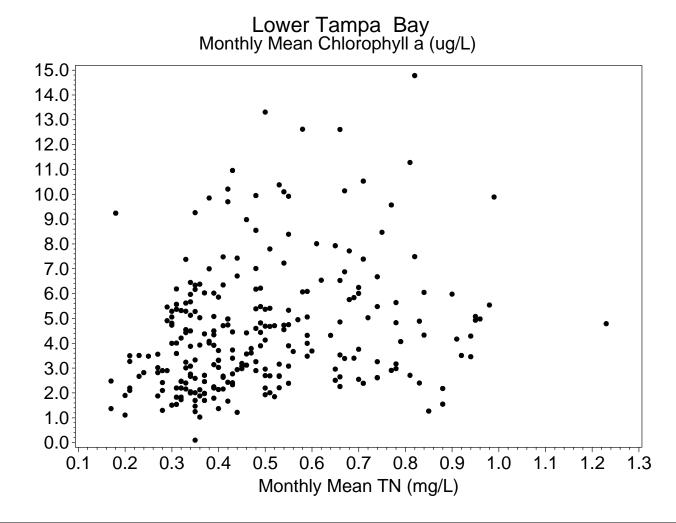


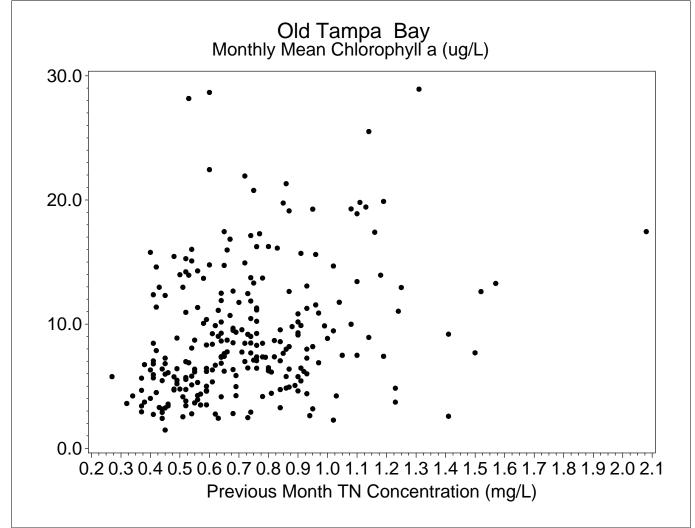
Attachment 5 Monthly Chlorophyll a Concentrations and Monthly TN Concentrations

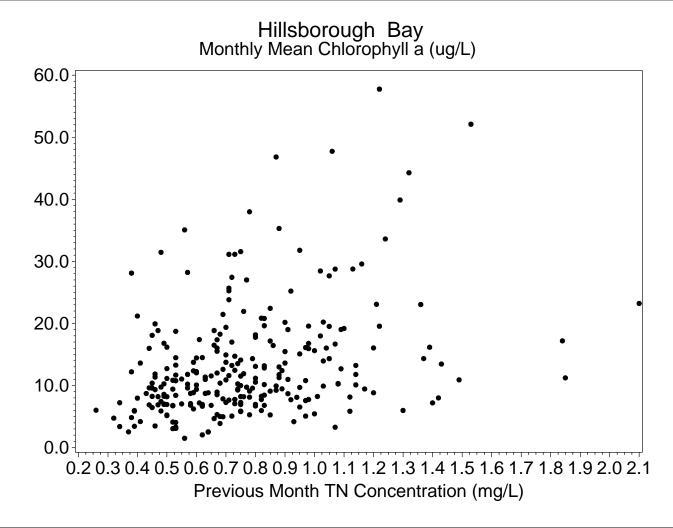


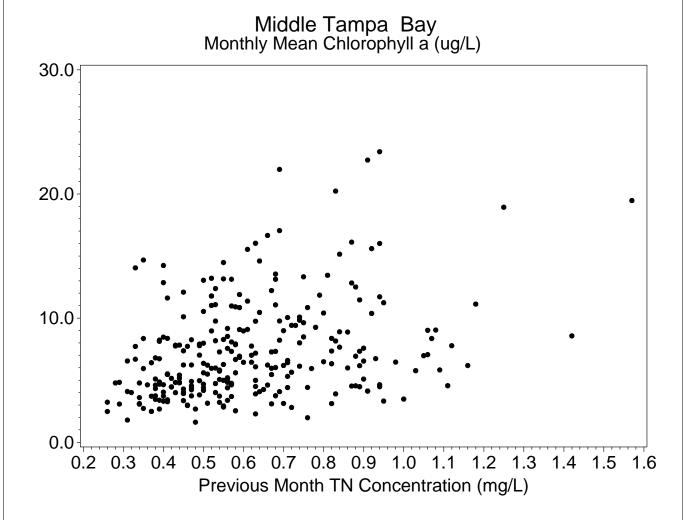


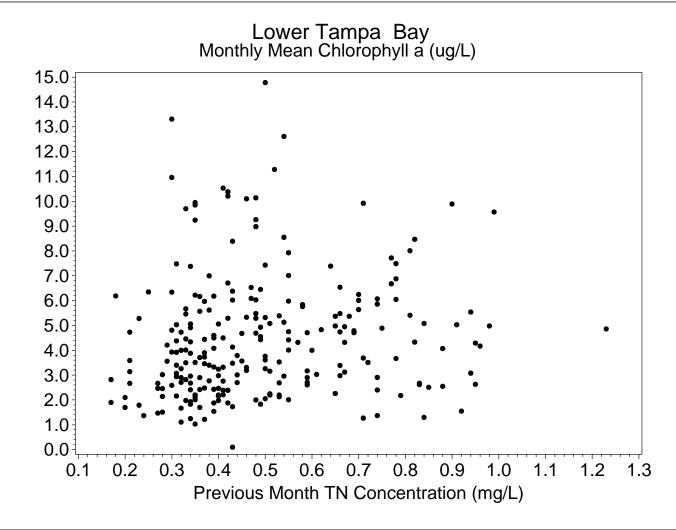


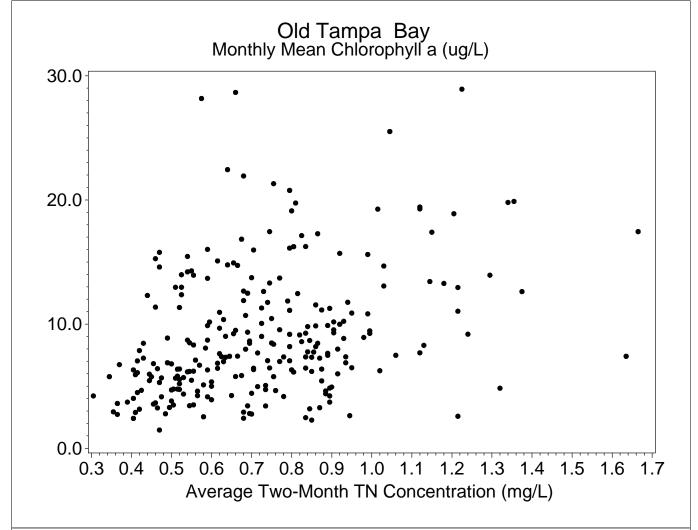


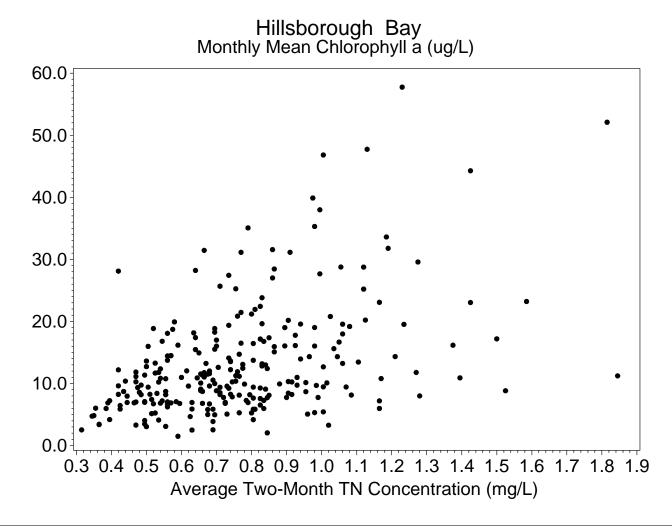


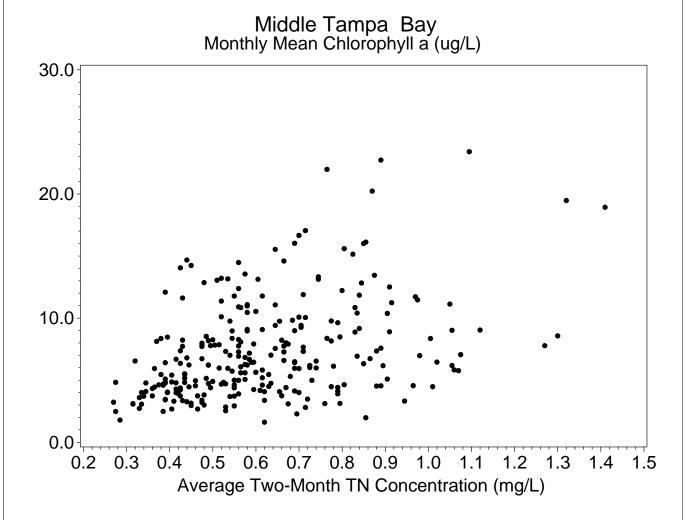


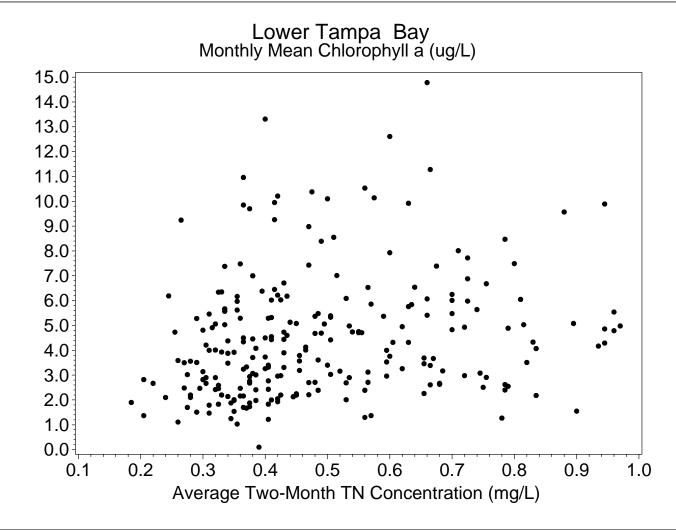


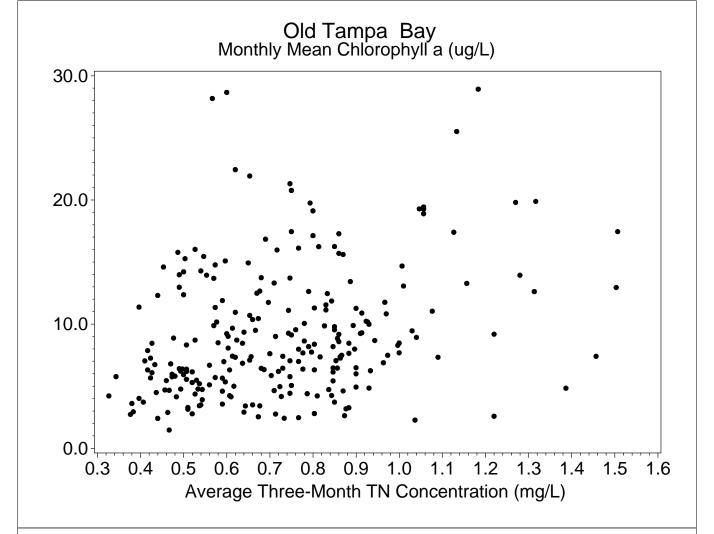


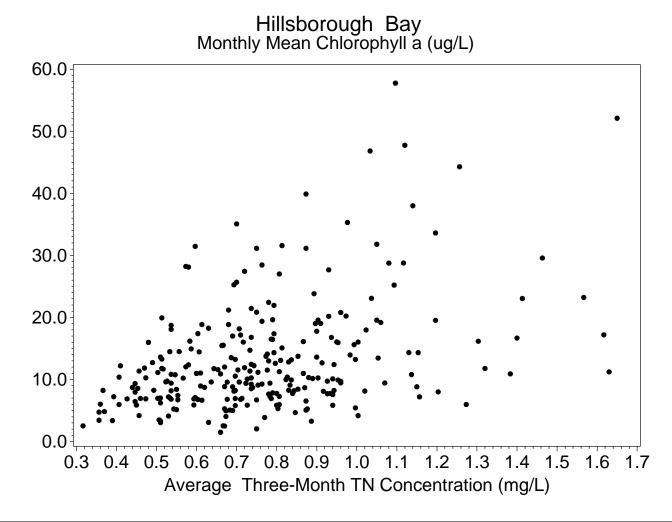


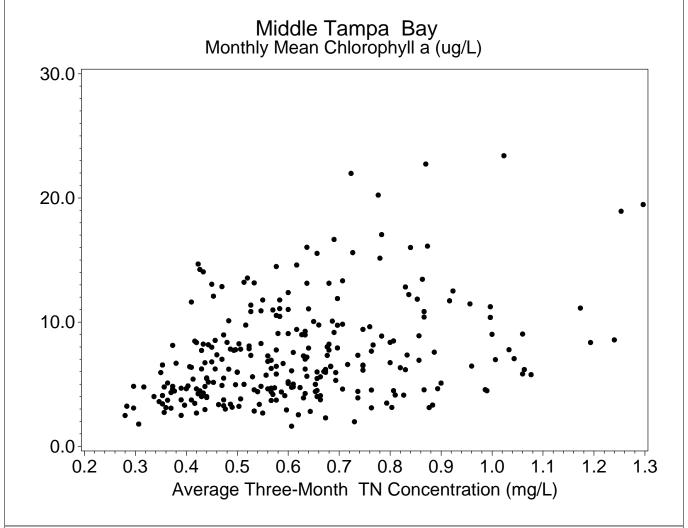


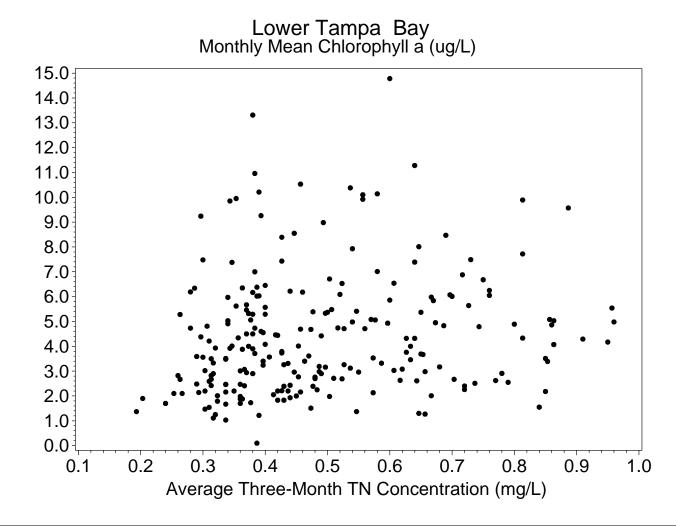












Attachment 6 Monthly Chlorophyll a Concentrations and Monthly TP Concentrations

