Robust and Powerful Trend Analyses for Continuous Water Quality Monitoring within the Matlacha Pass Aquatic Preserve (2005-2011)

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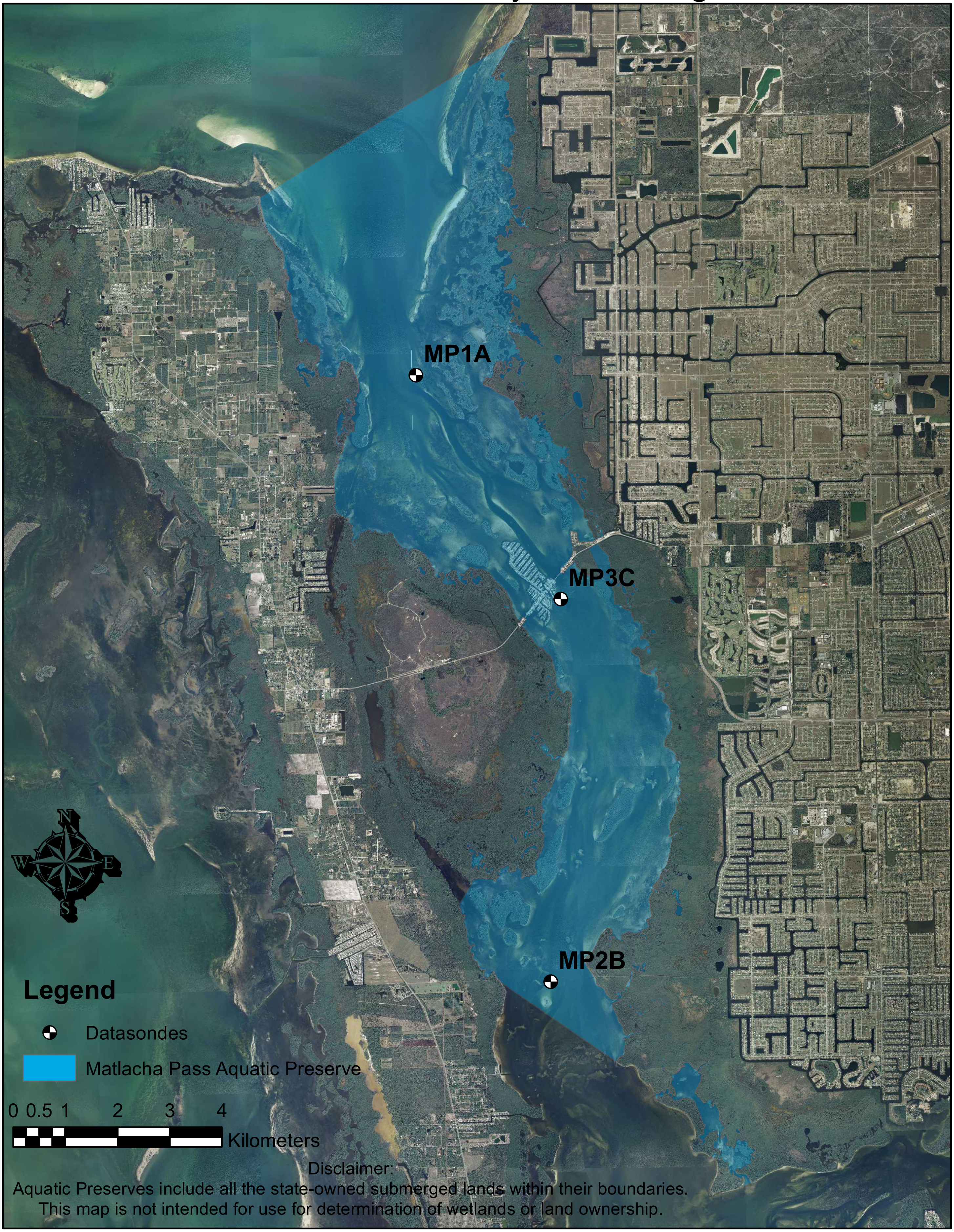
Matlacha Pass Aquatic Preserve Datasonde Continuous Water Quality Monitoring Sites



Abstract:

Since 2005, the Florida Department of Environmental Protection, Charlotte Harbor Aquatic Preserves have monitored water quality within the Matlacha Pass Aquatic Preserve using continuously deployed datasondes at two locations; one in northern Matlacha Pass and the other in southern Matlacha Pass. A third datasonde was deployed near the tidal node at the draw bridge in 2009. The program is intended to continuously track water quality status and trends within the preserve and relate them to influences from Charlotte Harbor to the north, the city of Cape Coral to the east, and the Caloosahatchee River to the southeast. Analyses of data from 2005 through 2011 show that dissolved oxygen, pH, total nitrogen, total Kjeldahl nitrogen, and total phosphorus were declining; and that chlorophyll a was increasing for Matlacha Pass. There were decreasing trends at MP1A for total nitrogen, total Kjeldahl nitrogen, ammonia, and total phosphorus; whereas chlorophyll a had increased. Site MP2B had significant decreases in dissolved oxygen, pH, total nitrogen and total Kjeldahl nitrogen, and chlorophyll a increased. When correcting for time-frame, site MP3C had the lowest dissolved oxygen, pH, and nutrients. Overall, site MP3C was more dissimilar from sites MP1A and MP2B than they were of each other. Continued monitoring is necessary in order to quantify trends at MP3C, relate water quality trends region-wide to seagrass trends, and to aid in the management of submerged resources within the region.

Introduction:

The Charlotte Harbor watershed, located in southwest Florida, drains into the vast estuarine complex of Charlotte Harbor which is one of the most pristine and socio-economically important ecosystems in the state. Located within this complex are many sub-estuaries, from Lemon Bay in the north to Estero Bay in the south, comprising nearly 250,000 acres of submerged habitat (e.g.: Duffey et al, 2007; Leary, 2009; and Leary 2010). Contained within the estuarine complex are six aquatic preserves. Aquatic preserves are areas of exceptional submerged resources that were set aside by the Florida legislature to be maintained in their essentially natural condition for future generations (Chapter 18-20, FAC). The aquatic preserves are managed by the FDEP Office of Coastal and Aquatic Managed Areas (CAMA). Five of the local aquatic preserves are managed by the Charlotte Harbor Aquatic Preserves' office in Punta Gorda, and include: Lemon Bay, Gasparilla Sound-Charlotte Harbor, Cape Haze, Pine Island Sound, and Matlacha Pass Aquatic Preserves. The sixth local aquatic preserve is the Estero Bay Aquatic Preserve, which is managed from the Estero Bay office located in Fort Myers Beach (Leary 2011a).

The purpose of the Matlacha Pass Datasonde Program is to collect continuous data in order to characterize water quality conditions within Matlacha Pass and relate it to influences from Charlotte Harbor to the north; the Cape Coral/Matlacha watershed to the east; and the lower Caloosahatchee River drainage to the south of the Pass. The program seeks to determine differences in water quality status and trends between individual sites and by season, in Matlacha Pass due to the location of the tidal node. In addition, to use the data with Charlotte Harbor Aquatic Preserves' Charlotte Harbor Estuaries Volunteer Water Quality Monitoring Network (CHEVWQMN) water quality data and seagrass transect monitoring data in order to focus resource management efforts and compliment other agencies water quality data and projects (Brown 2011).

The purpose of this report is to describe trends in water quality within the Matlacha Pass Aquatic Preserve for nearly six years of continuous water quality monitoring and monthly sampling (September 2005 – April 2011). The parameters of interest for trend analyses are data collected by datasondes including: dissolved oxygen (as a percentage of total saturation; DO), temperature, pH, turbidity, and salinity; as well as those collected monthly for laboratory analyses, including: chlorophyll *a*, total phosphorus (TP), total nitrogen (TN), ammonia, total Kjeldahl nitrogen (TKN), and nitrites + nitrates (NOx). These parameters are important in assessing the health of the estuary. This report summarizes water quality and relates it to location for the Matlacha Pass Aquatic Preserve.

Methods:

Study Area:

Situated in southwest Florida, the Matlacha Pass Aquatic Preserve is located within Lee County. The pass extends north to south between Pine Island to the west and the city of Cape Coral to the east, with the exception of the southern portion; the boundary of which follows the mean high water line. Its primary tributary is the Caloosahatchee River to the south. Matlacha Pass is approximately 21 km long and 5 km wide. The aquatic preserve encompasses approximately 14,600 acres of submerged habitat and mangrove islands. It is a shallow, micro-tidal bar-built sub-estuary of the greater Charlotte Harbor estuarine system. The Matlacha Pass Aquatic Preserve was established in 1972.

Field Methods:

Two datasondes were deployed in north and south Matlacha Pass during September 2005 in order to depict general open water conditions in real time. Station MP1A is in the northern part of Matlacha Pass, and is influenced by waters from Charlotte Harbor; and MP2B in the southern section which is primarily affected by waters from the Caloosahatchee River. A third site (MP3C) was established in March 2009 in central Matlacha Pass just south of the draw bridge near the tidal node for Matlacha Pass, and receives a mixture of waters from the other two sites, as well as runoff from the city of Cape Coral. The datasondes are encased in PVC housing with a lock top to prevent theft. Every two weeks to one month, the sondes are swapped out in order to control for drifting of data and probe fouling. The datasondes are located 0.5 m off the bottom and continually collect data every 15. Water quality samples were collected monthly at the three locations, also at 0.5 meter off the bottom. Seven parameters were measured with the sondes, including: temperature, salinity, conductivity, pH, DO, turbidity and depth. Additional monthly water quality samples included: TKN, NO_X, TP, ammonia, pheophytin *a* and chlorophyll *a*. Secchi depth and red tide and other HABs samples were also taken, but were not analyzed for this report (Brown 2011). In addition, pheophytin *a* and depth were not analyzed for this report.

Statistical Methods (adapted from Leary, 2011b):

Statistics were run using multiple methods on the following computer programs: SPSS 19.0 (IBM), Kendall.exe (USGS), KTRLine 1.0 (USGS), G*Power 3 (Faul et al 2007), and PAST 2.14 (Hammer et al 2001).

A priori power analyses were run based on Cohen's D and adjusted to R^2 in order to determine the minimum number of results (samples) required for statistical analyses (α=0.05; β= 0.05; and two tailed). It was determined that a minimum of 28 samples are required for regression analyses in order to detect a large effect size ($R^2 \geq 0.26$; e.g., Faul et al 2007, Cohen 1992, Lenth 2001, Lenth 2007 and Leech et al 2005). In addition, trend tests require a minimum of four to five years of data, depending on the specific analysis. Site MP3C was not analyzed for trends, because it has only been operational since January 2010, and data are only meant to give a very rough estimate of the status of this site; and to compare it with the other sites. Afterwards, *post hoc* power analyses were run to measure effect size. Exploratory/summary statistics were also run. Multiple tests were then run in order to test the assumption of normality, including: Johnson's SU transformation for skew; Anscombe & Glynn's transformation for kurtosis; and Jarque & Bera LM test (Jarque and Bera 1980, Jarque and Bera 1987 and DeCarlo 1997). Outliers were identified using Mahalanobis D^2 and by SARIMA (Seasonal Auto-Regressive Integrated Moving Average). The assumption of homoscedasticity was tested using the Breusch-Pagan test (Breusch and Pagan 1979).

From these statistics, assumptions of normality and homoscedasticity can be assessed; however since both parametric and non-parametric tests were run, the data were not transformed. In addition, assumptions of linearity were examined using plots of observed versus predicted values and residuals versus predicted values (e.g., Park 2008). Assumptions of independence were assessed using autocorrelation function (ACF) combined with the Durbin-Watson d test (e.g., Durbin and Watson 1950, Durbin and Watson 1951, Watson and Durbin 1951 and Dufour and Dagenais 1985). For time series analyses the assumption of stationarity (data are statistically in equilibrium, with constant mean and variance) was tested using ACF, Partial Auto-Correlation Function (PACF) and Ljung-Box Q; and is adjusted for by differencing in the model of the raw data, and is removed in the seasonally adjusted data (Box and Pierce 1970, Ljung and Box 1978, and Hamza 2009). In addition, using SARIMA, rather than ARIMA, allows for analyzing data multiplicatively by incorporating both a non-seasonal and a seasonal component (e.g., Durdu 2010).

Outliers that arise from obvious equipment malfunctions, transcription errors, or do not meet laboratory quality control criteria were excluded from analyses; or in the case of transcription errors corrected when identified (i.e., CDMO 2007, Small 2008 and YSI 2009). Outliers related to specific events (e.g., hurricanes, etc.) will be retained in the analyses. Laboratory data reported as below the method detection limit (MDL) will be analyzed as the maximum MDL for that parameter, as well as all values reported below the maximum MDL. Although this substitution method overestimates the data, it was used due to its ease and speed (e.g., Helsel 1990 and Helsel 2010). In order to better handle such a large dataset with multiple cyclical components (daily, tidal, annual, etc.), monthly means and medians were used for analyses (e.g., Burkholder et al 2006). Missing data were re-coded for time series analyses as the mean or median for that parameter for that particular month. Raw data was adjusted using Seasonal Decomposition, which results in three data sets: the original "raw" data; seasonally adjusted data (data that have annual cyclical patterns removed; SAS); and trend-cycle smoothed data (a smoothed version of the seasonally adjusted data; STC). All data were assumed to have at least an annual periodicity for Seasonal Decomposition.

Multiple tests (parametric, robust, and non-parametric) were used to detect trends, including: Seasonal Kendall Trend Test (SKTT; Hirsch et al 1982, Hirsch and Slack 1984, Hirsch et al 1991, Helsel and Hirsch 2002, and Helsel et al 2006); Theil-Kendall robust line regression (KTRL; Theil 1950a, Theil 1950b, Theil 1950c, Granato 2006 and Sen and Saleh 2010); SARIMA models; exponential smoothing models (these and SARIMA are collectively called time series models); Robust Regression (based on M-estimators, rather than mean or median; RR; e.g., Nevitt and Tam 1998); and Ordinary Least Squares regression (OLS; e.g., Burkholder et al 2006, and Weber and Perry 2006). All models, with the exception of SKTT and KTRL, were run on the raw data; SAS data; and STC data. However, only results for those models where assumptions were met are presented. **Note**: SPSS Time Series Modeler includes a procedure called Expert Modeler, which identifies the best fitting SARIMA or exponential smoothing model, was used rather than modeling the parameters by trial and error; in addition this procedure automatically transforms (either square root or natural log) data when necessary (SPSS 2010). Trends in time series models were obtained by using the Least Squares method on the predicted (modeled) values (Burkholder et al 2006). Measures of goodness of fit include: stationary R^2, R^2, root mean square error (RMSE), mean absolute percent error (MAPE), maximum absolute percent error (MaxAPE), mean absolute error (MAE), normalized Bayesian information criterion (BIC), median deviation, median absolute deviation (MAD), non-parametric prediction error sum of squares (NPPRESS), and bias correction factor (BCF). Both t-tests and F-tests were performed on the observed versus the predicted values in the time series analyses in order to determine if they were statistically significantly different.

When statistical assumptions are met, parametric statistics (based on metrics; for example mean, median, standard deviation, etc.) offer the best power and accuracy, but if they are violated the error components of the analyses become overly inflated and the statistics become unreliable (e.g., Yaffee 2002). When this occurs, it becomes necessary to employee either robust statistics (based on estimators) or nonparametric statistics (based on ranks; Erceg-Hum and Mirosevich 2008). Although robust statistics can improve power and accuracy when assumptions are violated, there are no set standards for when the data are too non-normal, heteroscedastic, or when outliers are too influential. Therefore nonparametric statistics were employed, as well. These multiple methods allow for better interpretation of results (Wilcox 2001).

When data do not meet specified parametric assumptions of normality or homoscedasticity, transformations can be applied to the data in order to run parametric statistics. Typical transformations include: logarithmic, square root, exponential and reciprocal. For reciprocal, Box-Cox and log transformations, the data must not equal zero (e.g., Yamamura 1999). Although transformations can help to stabilized variance, and in the process aid in producing a normal distribution, there are instances where transformations will not satisfy assumptions. These include: heavy tailed distributions; bimodal or multimodal distributions; distributions with a large number of equal values; and where transformations lead to variation in the p-value. In addition, transformations can only be used on discrete or continuous data (e.g., Yaffee 2002 and Helal 2008). When assumptions are not met and transformations do not help or cannot be used, non-parametric statistics or robust statistics must be used. In addition, transformations can change the underlying nature of the data (Osborne 2002). Therefore, for this paper, transformations were not used, with the exception of SARIMA models where SPSS Expert Modeler did so.

Where data were normal ANOVA, Levene's, Welch F, and Gabriel's tests were used for site comparisons, and the mean was reported. However, if data were non-normal, Kruskal-Wallis H and Man-Whitney U were used for site comparisons, and the median was reported.

Results are given for the Matlacha Pass Aquatic Preserve as a whole and by site. For detailed statistics, see the appendices contained within Leary, 2011b.

Results:

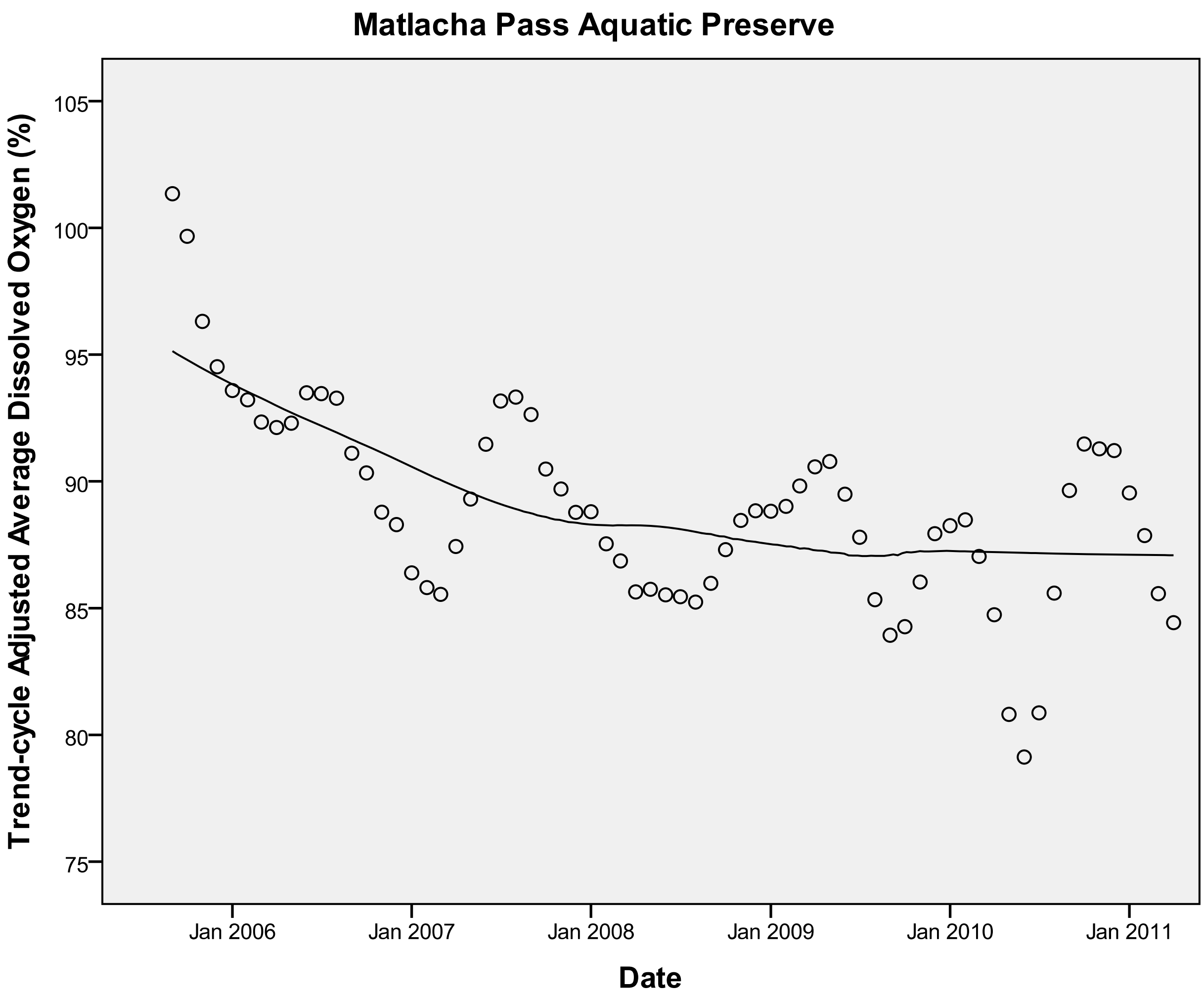
Field Parameters

Dissolved Oxygen

Statistical assumptions were violated for all DO data within the Matlacha Pass Aquatic preserve, therefore nonparametric results were used. Both the SKTT and KTRL results showed significant ($p<0.05$ for both) declining trends in DO at rates between -1.1% and -1.2% per year, respectively. In addition, Matlacha Pass Aquatic Preserve's SARIMA models of the SAS and STC DO showed significant decreasing trends in DO; the OLS regressions of the SAS and STC data; and the RR regressions of the SAS and STC data ($p<0.05$ for all trends and models). Regressions, both OLS and RR, of the raw data were not significant, as was the SARIMA model of the raw data. The same general pattern was observed for site MP2B, as well; with significant declining trends in most models. Both the OLS and RR regressions of the seasonally adjusted data did not violate assumptions and were decreasing significantly ($p<0.05$ for both) at rates of approximately -1% per year for MP2B. However, unlike the Aquatic Preserve, the non-parametric tests were not significant. The only significant result for MP1A was for RR regression of the STC data (increasing; $p=0.035$), however all assumptions were violated. There were no other significant trends in DO. Site MP3c DO was significantly lower than the other two sites (mean DO: MP3C=80%, MP1A=90%, MP2B=91%; $p=0.001$). The mean DO for Matlacha Pass Aquatic Preserve was 87% (95% CI=85 – 90%).

DO (% Saturation)								
Matlacha Pass Aquatic Preserve			MP1A			MP2B		
Model or Trend Test	Slope	P	Model or Trend Test	Slope	P	Model or Trend Test	Slope	P
DO_SARIMA(0,0,1)(1,0,0)$_{12}$	-0.58	0.162	DO_Simple Seasonal	-0.17	0.770	DO_SARIMA(1,0,0)(0,1,0)$_{12}$	0.19	0.808
DO-SAS_SARIMA(1,0,0)(0,0,0)$_{12}$	**-0.70**	**0.000**	DO-SAS_SARIMA(0,0,2)(0,0,0)$_{12}$	0.02	0.929	DO-SAS_Simple Seasonal	**-1.04**	**0.001**
DO-STC_SARIMA(1,1,0)(0,0,0)$_{12}$	**-1.34**	**0.000**	DO-STC_SARIMA(1,1,0)(1,0,0)$_{12}$	0.08	0.820	DO-STC_Damped Trend	**-0.90**	**0.004**
DO_OLS	-0.76	0.174	DO_OLS	0.53	0.393	DO_OLS	-0.36	0.524
DO-SAS_OLS	**-1.54**	**0.000**	DO-SAS_OLS	-0.16	0.731	DO-SAS_OLS	**-1.07**	**0.014**
DO-STC_OLS	**-1.46**	**0.000**	DO-STC_OLS	-0.12	0.724	DO-STC_OLS	**-0.96**	**0.001**
DO_RR	-0.70	0.234	DO_RR	0.51	0.438	DO_RR	-0.35	0.533
DO-SAS_RR	**-1.38**	**0.000**	DO-SAS_RR	0.16	0.728	DO-SAS_RR	**-0.89**	**0.025**
DO-STC_RR	**-1.38**	**0.000**	DO-STC_RR	**0.63**	**0.035**	DO-STC_RR	**-0.98**	**0.001**
DO_SKTT	**-1.08**	**0.001**	DO_SKTT	-0.12	0.907	DO_SKTT	-0.58	0.102
DO_KTRL	**-1.21**	**S**	DO_KTRL	0.38	NS	DO_KTRL	-0.76	NS

Note: KTRLine does not report P-values, therefore if the 95% CI did not contain zero it was considered significant (S).



Temperature

Statistical assumptions were violated for all temperature data within the Matlacha Pass Aquatic preserve, therefore nonparametric results were used. The only significant trend in temperature for the Matlacha Pass Aquatic Preserve violated assumptions and was a decreasing trend in the RR regression of the STC data. Site MP1A also had a significant decreasing trend in the OLS regression of the STC data; and the SARIMA predictive model of the STC data had a significant decreasing trend, however these too violated assumptions. As with the Aquatic Preserve, MP2B had a significant decreasing trend in temperature for the RR regression of the STC data, but violated assumptions. However, *post hoc* analyses were run because the data appeared to be bimodal. A two-step cluster analysis was performed on the data and showed three clusters (seasons), one centered around a mean of 29.87°C for the months of May through September (summer); one centered around a mean of 23.90°C for the months of March through April and October through November (transitional; spring and fall); and the last centered around a mean of 18.80°C for December through February (winter). When assumptions were tested on the three clusters, none were violated at the 0.05 level. Regressions of the three clusters showed that the summer values were increasing ($p=0.048$) and the winter values were decreasing ($p=0.003$). The mean temperature for the aquatic preserve was 24.83°C (95% CI=23.62 – 26.03°C). There were no other significant trends in temperature, nor were there significant differences between the sites.

Clusters

Input (Predictor) Importance

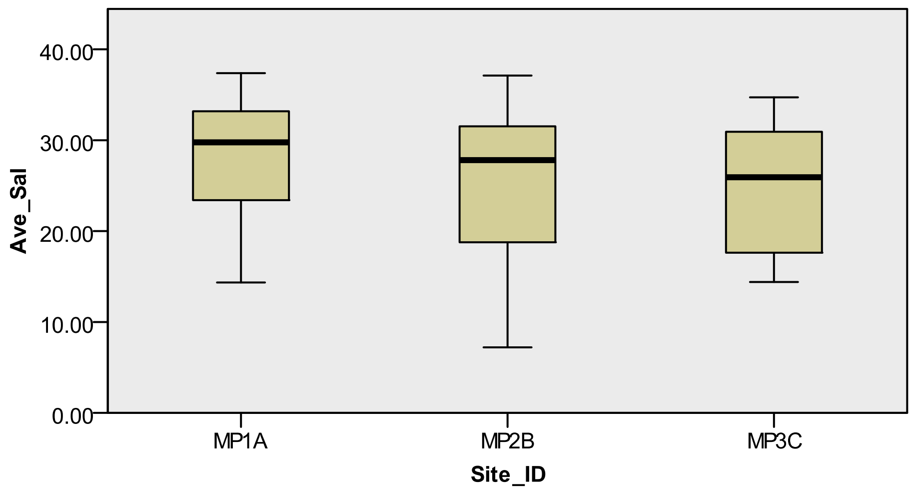
1.0 0.8 0.6 0.4 0.2 0.0

Cluster	1	2	3
Label			
Description			
Size	38.3% (62)	35.8% (58)	25.9% (42)
Inputs	Ave_T 29.87	Ave_T 23.90	Ave_T 18.80
	Month 9 (22.6%)	Month 3 (25.9%)	Month 12 (33.3%)

Salinity

There were no significant trends in salinity for the Matlacha Pass Aquatic Preserve or for the individual sites. The mean salinity for Matlacha Pass Aquatic Preserve was 25.66 PSU (95%CI=24.25 – 27.07 PSU).

Independent-Samples Kruskal-Wallis Test



Total N	162
Test Statistic	6.390
Degrees of Freedom	2
Asymptotic Sig. (2-sided test)	.041

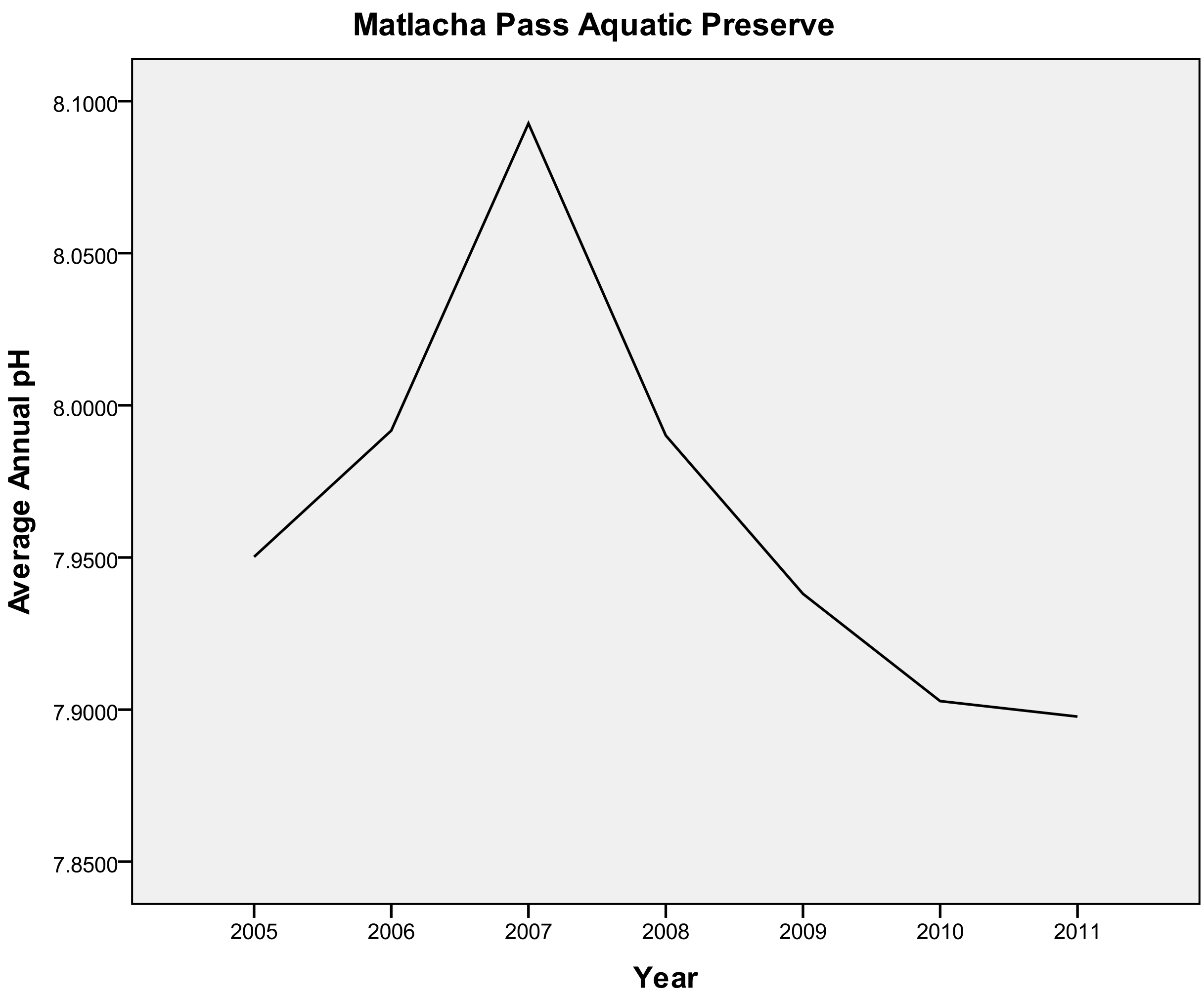
1. The test statistic is adjusted for ties.

pH

Statistical assumptions were violated for all pH data within the Matlacha Pass Aquatic preserve, therefore nonparametric results were used. All trend analyses and models of pH for the Matlacha Pass Aquatic Preserve showed significant decreasing trends. The nonparametric results showed pH to be decreasing at approximately -0.025 SU per year ($p<0.05$ for both). However, site MP1A only had significant decreasing trends in the regressions (both OLS and RR) of the SAS and STC pH data; and a significant decreasing trend in the SARIMA of the STC data, however assumptions were violated. Site MP2B had significant decreasing trend for pH for all parametric regressions and SARIMA models of the SAS and STC data, however only the SAS data did not violate assumptions. There were no other significant trends for pH. Site MP3C was significantly ($p<0.001$) lower than the other sites (median pH: MP3C=7.77, MP2B=7.93, and MP1A=8.01). The median pH for the aquatic preserve was 7.91.

pH								
Matlacha Pass Aquatic Preserve			**MP1A**			**MP2B**		
Model or Trend Test	Slope	P	Model or Trend Test	Slope	P	Model or Trend Test	Slope	P
pH_SARIMA(1,0,0)(0,0,0)$_{12}$	**-0.02**	**0.014**	pH_SARIMA(0,0,0)(1,1,0)$_{12}$	-0.02	0.312	pH_SARIMA(0,0,1)(0,0,0)$_{12}$	-0.01	0.074
pH-SAS_SARIMA(0,0,0)(0,0,0)$_{12}$	**-0.02**	**0.004**	pH-SAS_SARIMA(0,0,0)(0,0,1)$_{12}$	-0.04	0.060	pH-SAS_SARIMA(0,0,1)(0,0,0)$_{12}$	**-0.01**	**0.040**
pH-STC_SARIMA(0,1,1)(0,0,0)$_{12}$	**-0.04**	**0.000**	pH-STC_SARIMA(0,0,2)(0,0,0)$_{12}$	**-0.02**	**0.000**	pH-STC_SARIMA(0,1,3)(0,0,0)$_{12}$	**-0.02**	**0.000**
pH_OLS	**-0.02**	**0.028**	pH_OLS	-0.03	0.156	pH_OLS	-0.02	0.135
pH-SAS_OLS	**-0.03**	**0.001**	pH-SAS_OLS	**-0.04**	**0.047**	pH-SAS_OLS	**-0.02**	**0.006**
pH-STC_OLS	**-0.03**	**0.000**	pH-STC_OLS	**-0.03**	**0.000**	pH-STC_OLS	**-0.02**	**0.000**
pH_RR	**-0.02**	**0.045**	pH_RR	-0.01	0.392	pH_RR	-0.01	0.304
pH-SAS_RR	**-0.03**	**0.000**	pH-SAS_RR	**-0.02**	**0.013**	pH-SAS_RR	**-0.02**	**0.023**
pH-STC_RR	**-0.03**	**0.000**	pH-STC_RR	**-0.02**	**0.002**	pH-STC_RR	**-0.02**	**0.000**
pH_SKTT	**-0.02**	**0.022**	pH_SKTT	-0.02	0.175	pH_SKTT	-0.02	0.112
pH_KTRL	**-0.03**	**S**	pH_KTRL	-0.01	NS	pH_KTRL	-0.02	NS

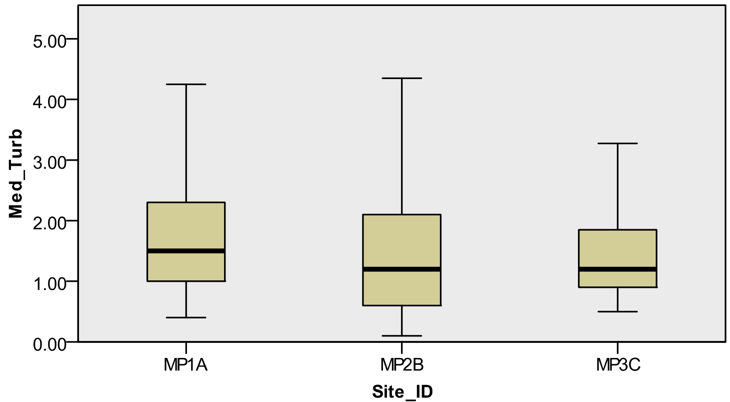
Note: KTRLine does not report P-values, therefore if the 95% CI did not contain zero it was considered significant (S).



Turbidity

Statistical assumptions were markedly violated for all turbidity data within the Matlacha Pass Aquatic preserve, therefore nonparametric results were used. The SARIMA model for the Matlacha Pass Aquatic Preserve of the STC turbidity data was significantly decreasing, as were the OLS and RR regressions of the STC data and the OLS regression of the SAS data. There were significant decreasing trends of the OLS regressions of the SAS and STC turbidity data for MP2B. This site also had significant decreasing trends in the SARIMA models of the raw data and the STC data. All significant trend tests and models violated statistical assumptions for the aquatic preserve as a whole, as well as for the individual sites. There were no other significant turbidity trends, nor was there a significant difference between the median values for the sites. The median turbidity for the aquatic preserve was 2.21 NTU.

Independent-Samples Kruskal-Wallis Test



Total N	158
Test Statistic	4.293
Degrees of Freedom	2
Asymptotic Sig. (2-sided test)	.117

1. The test statistic is adjusted for ties.
2. Multiple comparisons are not performed because the overall test does not show significant differences across samples.

Laboratory Parameters

TN

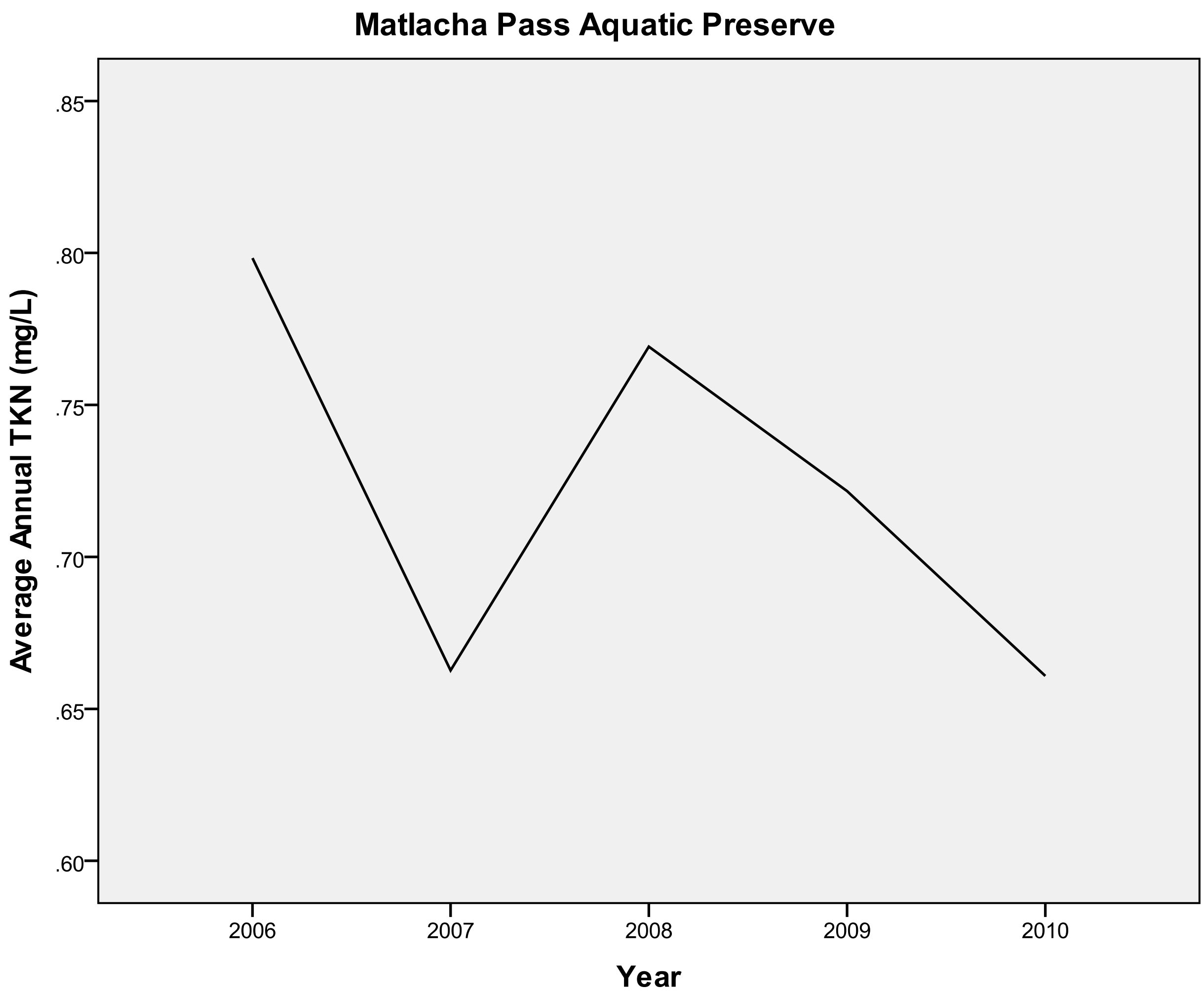
Statistical assumptions, particularly normality, were markedly violated for all TN data within the Matlacha Pass Aquatic preserve, therefore nonparametric results were used. There were significant declining trends in TN for all models and trend analyses for the Matlacha Pass Aquatic preserve, with the exception of the SARIMA model of the raw data and the SKTT. The KTRL nonparametric analysis showed TN to be declining at a rate of -0.03 mg/L per year ($p<0.05$). All models and trend analyses of TN were significantly decreasing for site MP1A; however assumptions of normality were violated. Both nonparametric trend tests showed TN to be decreasing at a rate of -0.04 mg/L per year at MP1A ($p<0.05$). And, site MP2B had significantly decreasing trends and models for TN, with the exception of the SARIMA model of the raw data. Although assumptions were violated at MP2B, nonparametric tests confirmed that TN was declining significantly ($p<0.05$) at MP2B at an approximate rate of -0.04 mg/L per year. There were no other significant trends in TN. There was no significant difference between MP1A and MP2B with regards to median TN, whereas MP3C was significantly lower than the other two locations (median TN: MP3C=0.41, MP2B=0.62, and MP1A=0.64 mg/L; p=0.021). The median TN for Matlacha Pass Aquatic Preserve was 0.56 mg/L.

TN								
Matlacha Pass Aquatic Preserve			**MP1A**			**MP2B**		
Model or Trend Test	Slope	P	Model or Trend Test	Slope	P	Model or Trend Test	Slope	P
$SARIMA(1,0,0)(0,0,0)_{12}$	-0.02	0.119	$SARIMA(0,0,0)(0,0,0)_{12}$	**-0.03**	**0.000**	$SARIMA(1,0,0)(0,0,0)_{12}$	-0.02	0.139
SAS_Simple	**-0.05**	**0.000**	$SAS\_SARIMA(0,0,0)(0,0,0)_{12}$	**-0.03**	**0.000**	$SAS\_SARIMA(1,0,0)(0,0,0)_{12}$	-0.02	0.021
$STC\_SARIMA(0,1,2)(0,0,0)_{12}$	**-0.04**	**0.000**	$STC\_SARIMA(0,1,3)(0,0,0)_{12}$	**-0.04**	**0.000**	$STC\_SARIMA(0,1,3)(0,0,0)_{12}$	-0.05	0.000
OLS	**-0.04**	**0.018**	OLS	**-0.04**	**0.005**	OLS	-0.05	0.023
SAS_OLS	**-0.04**	**0.006**	SAS_OLS	**-0.04**	**0.004**	SAS_OLS	-0.05	0.017
STC_OLS	**-0.04**	**0.000**	STC_OLS	**-0.04**	**0.000**	STC_OLS	-0.05	0.001
RR	**-0.03**	**0.006**	RR	**-0.04**	**0.001**	RR	-0.04	0.015
SAS_RR	**-0.02**	**0.049**	SAS_RR	**-0.03**	**0.001**	SAS_RR	-0.03	0.016
STC_RR	**-0.02**	**0.005**	STC_RR	**-0.03**	**0.000**	STC_RR	-0.03	0.009
SKTT	-0.03	0.052	SKTT	**-0.04**	**0.020**	SKTT	-0.04	0.018
KTRL	**-0.03**	**S**	KTRL	**-0.04**	**S**	KTRL	-0.04	S

Note: KTRLine does not report P-values, therefore if the 95% CI did not contain zero it was considered significant (S).

TKN

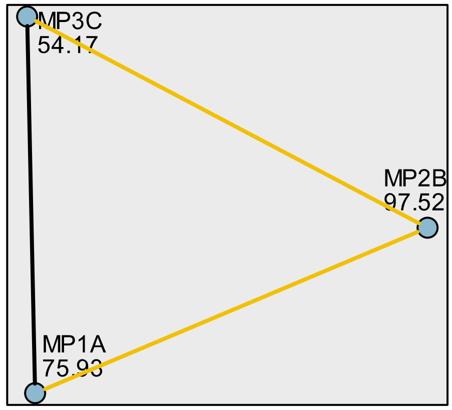
Statistical assumptions were violated for all TKN data within the Matlacha Pass Aquatic preserve and at both sites, therefore nonparametric results were used. All models and trend analyses of TKN were significantly decreasing for the Aquatic Preserve and the two sites, with the exception of the SARIMA model of the raw data of the entire Aquatic Preserve and the SARIMA models of the raw data and the SAS data at MP2B. Nonparametric trend analyses showed TKN to be declining in the aquatic preserve at a rate of -0.03 mg/L per year ($p<0.05$ for both). Site MP1A TKN was trending down at a slightly faster rate of -0.04 mg/L per year ($p<0.05$ for both SKTT and KTRL). And, MP2B was decreasing at approximately -0.035 mg/L per year ($p<0.05$ for both SKTT and KTRL). There were no other significant trends in TKN. There was no significant difference between MP1A and MP2B with regards to median TKN, whereas MP3C was significantly lower than the other two locations (median TKN: MP3C=0.40, MP2B=0.60, and MP1A=0.63 mg/L; p=0.022). The median TKN for the aquatic preserve was 0.54 mg/L.



Ammonia

Statistical assumptions were violated for all ammonia data within the Matlacha Pass Aquatic preserve and at both sites, therefore nonparametric results were used. There were no significant trends in ammonia for the Matlacha Pass Aquatic Preserve as a whole. However, there were significant declining trends in ammonia for the SKTT, RR regressions of the SAS and STC data, and the SARIMA model at site MP1A. The SKTT for MP1A was declining at a rate of less than -0.001 mg/L per year (p=0.049). And, there were significant increasing trends at site MP2B for the OLS regression of the STC data and the SARMIA model of the STC data, but as stated earlier, assumptions were violated. There were no other significant trends in ammonia. There was no significant difference between MP1A and MP3C with regards to median ammonia, whereas MP2B was significantly higher than the other two locations (median ammonia: MP3C=0.01, MP1A=0.02, and MP2B=0.03 mg/L; p=0.008). For the region as a whole, the median ammonia value was 0.02 mg/L.

Pairwise Comparisons of Site_ID



Each node shows the sample average rank of Site_ID.

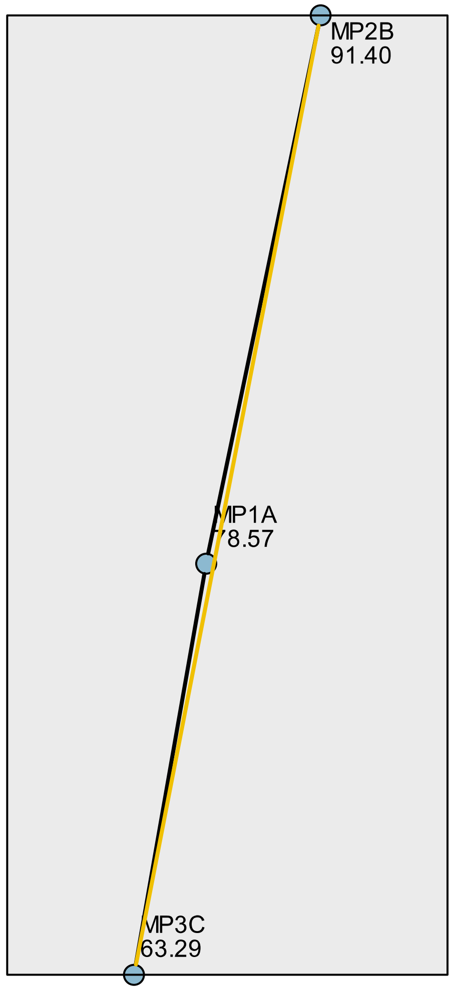
Sample 1-Sam...	Test Statistic	Std. Error	Std. Test Statistic	Sig.	Adj.Sig.
2-0	21.753	10.582	2.056	.040	.119
2-1	43.349	10.582	4.097	.000	.000
0-1	-21.596	7.870	-2.744	.006	.018

Each row tests the null hypothesis that the Sample 1 and Sample 2 distributions are the same.
Asymptotic significances (2-sided tests) are displayed. The significance level is .05.

NO_x

Statistical assumptions were violated for all NO_x data within the Matlacha Pass Aquatic preserve and at both sites, therefore nonparametric results were used. The Matlacha Pass Aquatic Preserve had significant increasing trends in NO_x for the RR regressions of the raw data and the STC data. However, the SARIMA model of the raw data for the Aquatic Preserve showed a declining trend. The RR regression of the STC data at site MP2B was increasing significantly. Although statistical assumptions were violated there was good agreement between robust methods and nonparametric methods, all of which showed increases in NO_x. There were no other significant trends for NO_x. There was no significant difference between MP1A and the other two sites with regards to median NO_x, whereas MP3C was significantly lower than MP2B (median NO_x: MP3C=0.006, MP1A=0.010, and MP2B=0.023 mg/L; p=0.043). The median value for NO_x for the aquatic preserve was 0.013 mg/L.

Pairwise Comparisons of Site_ID



Each node shows the sample average rank of Site_ID.

Sample 1-Sam...	Test Statistic	Std. Error	Std. Test Statistic	Sig.	Adj.Sig.
2-0	15.278	10.538	1.450	.147	.441
2-1	28.109	10.538	2.667	.008	.023
0-1	-12.831	7.838	-1.637	.102	.305

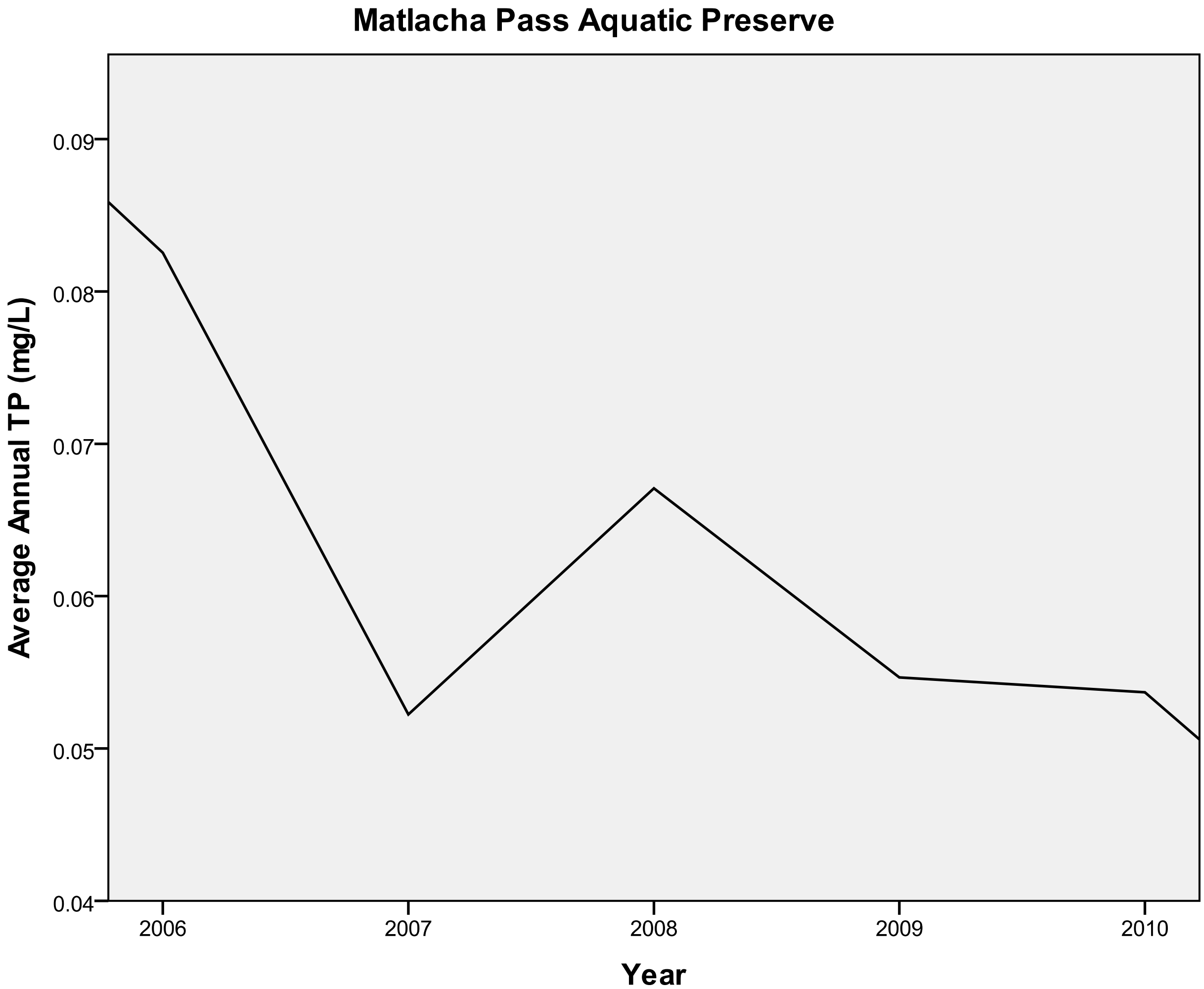
Each row tests the null hypothesis that the Sample 1 and Sample 2 distributions are the same.
Asymptotic significances (2-sided tests) are displayed. The significance level is .05.

TP

Statistical assumptions were violated for all TP data within the Matlacha Pass Aquatic preserve and at both sites, therefore nonparametric results were used. All trend analyses and SARIMA models for both the Matlacha Pass Aquatic Preserve and site MP1A were significantly declining. The SKTT and KTRL for the aquatic preserve had TP decreasing at a rate of between -0.004 and -0.005 mg/L per year ($p<0.05$ for both). Whereas, MP1A nonparametric analyses showed TP to be declining at a rate between -0.005 and -0.006 mg/L per year ($p<0.05$ for both). However, at site MP2B, the only significant trends were for the RR regressions of the SAS and STC data; the OLS regression of the STC data; and the SARIMA model of the STC data, each of which were trending down, but violated assumptions. There were no other significant trends in TP. There was no significant difference between MP1A and MP2B with regards to median TP, whereas MP3C was significantly lower than the other two locations (median TP: MP3C=0.026, MP2B=0.054, and MP1A=0.055 mg/L; $p<0.001$). The median value of TP for Matlacha Pass Aquatic Preserve was 0.045 mg/L.

TP								
Matlacha Pass Aquatic Preserve			**MP1A**			**MP2B**		
Model or Trend Test	Slope	P	Model or Trend Test	Slope	P	Model or Trend Test	Slope	P
$SARIMA(1,0,0)(0,0,0)_{12}$	**-0.002**	**0.019**	Winter's Multiplicative	**-0.006**	**0.000**	$SARIMA(0,0,1)(0,0,0)_{12}$	-0.001	0.500
$SAS\_SARIMA(0,1,1)(0,0,0)_{12}$	**-0.007**	**0.000**	$SAS\_SARIMA(1,0,1)(0,0,0)_{12}$	**-0.006**	**0.000**	$SAS\_SARIMA(0,0,1)(0,0,0)_{12}$	-0.001	0.626
$STC\_SARIMA(0,1,3)(0,0,0)_{12}$	**-0.006**	**0.000**	$STC\_SARIMA(0,1,3)(0,0,0)_{12}$	**-0.006**	**0.000**	$STC\_SARIMA(0,1,3)(0,0,0)_{12}$	**-0.004**	**0.000**
OLS	**-0.007**	**0.001**	OLS	**-0.007**	**0.000**	OLS	-0.005	0.071
SAS_OLS	**-0.006**	**0.000**	SAS_OLS	**-0.006**	**0.000**	SAS_OLS	-0.004	0.063
STC_OLS	**-0.006**	**0.000**	STC_OLS	**-0.006**	**0.000**	STC_OLS	**-0.003**	**0.001**
RR	**-0.005**	**0.001**	RR	**-0.005**	**0.002**	RR	-0.003	0.071
SAS_RR	**-0.005**	**0.000**	SAS_RR	**-0.006**	**0.000**	SAS_RR	**-0.003**	**0.024**
STC_RR	**-0.006**	**0.000**	STC_RR	**-0.006**	**0.000**	STC_RR	**-0.004**	**0.000**
SKTT	**-0.025**	**0.001**	SKTT	**-0.006**	**0.001**	SKTT	-0.003	0.052
KTRL	**-0.031**	**S**	KTRL	**-0.005**	**S**	KTRL	-0.002	NS

Note: KTRLine does not report P-values, therefore if the 95% CI did not contain zero it was considered significant (S).



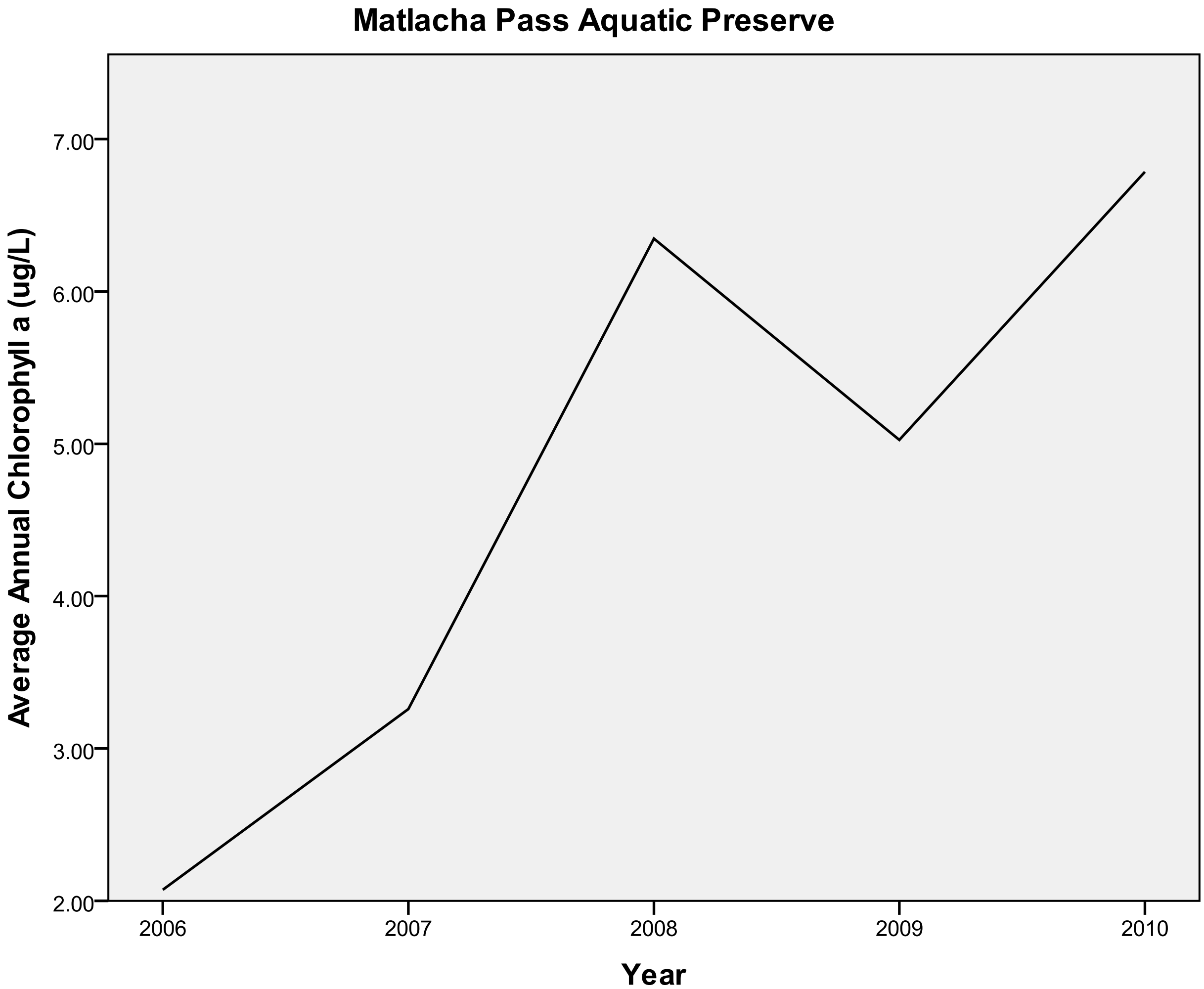
Chlorophyll *a*

With the exception of SAS data for the aquatic preserve, statistical assumptions were violated for all chlorophyll *a* data within the Matlacha Pass Aquatic preserve and at both sites, therefore nonparametric results were used (except for SAS results for the aquatic preserve). All models and trend analyses of chlorophyll *a* were significantly increasing for the Matlacha Pass Aquatic Preserve and the two sites, with the exception of the SARIMA models of the raw data at the two individual sites. Parametric results for chlorophyll *a* SAS trends in Matlacha Pass Aquatic Preserve (OLS and SARIMA) showed increases of 1.02 and 1.03 µg/L per year ($p<0.001$ for both); robust results were increasing at 1.06 µg/L per year ($p<0.001$); and nonparametric results increased at a rate of 0.66 and 0.70 µg/L per year for SKTT and KTRL, respectively ($p<0.05$ for both). Site MP1A had increasing trends for SKTT and KTRL at rates of 0.96 and 0.93 µg/L per year ($p<0.05$ for both). And, MP2B increased at a rate of 0.32 and 0.37 µg/L per year for SKTT and KTRL ($p<0.05$ for both). There were no other significant trends in chlorophyll *a*. There was no significant difference between MP3C and the other two sites with regards to median chlorophyll *a*, whereas MP2B was significantly lower than MP1A (median chlorophyll *a*:

MP2B=3.36, MP3C=4.30, and MP1A=7.03 µg/L; p=0.014). Matlacha Pass Aquatic Preserve had a median chlorophyll *a* concentration of 4.90 µg/L.

Chlorophyll a								
Matlacha Pass Aquatic Preserve			**MP1A**			**MP2B**		
Model or Trend Test	Slope	P	Model or Trend Test	Slope	P	Model or Trend Test	Slope	P
SARIMA(0,0,0)(1,0,0)$_{12}$	**0.74**	**0.000**	SARIMA(0,0,0)(0,1,1)$_{12}$	0.77	0.106	SARIMA(1,0,0)(0,0,0)$_{12}$	0.12	0.680
SAS_SARIMA(1,0,0)(0,0,0)$_{12}$	**1.03**	**0.000**	SAS_SARIMA(0,0,0)(0,0,0)$_{12}$	**1.30**	**0.000**	SAS_SARIMA(1,0,0)(0,0,0)$_{12}$	**0.30**	**0.031**
STC_SARIMA(0,1,3)(0,0,0)$_{12}$	**1.09**	**0.000**	STC_SARIMA(0,1,3)(0,0,0)$_{12}$	**1.63**	**0.000**	STC_SARIMA(0,1,1)(0,0,0)$_{12}$	**0.64**	**0.000**
OLS	**0.80**	**0.015**	OLS	**1.19**	**0.011**	OLS	0.19	0.575
SAS_OLS	**1.02**	**0.000**	SAS_OLS	**1.55**	**0.000**	SAS_OLS	**0.54**	**0.003**
STC_OLS	**1.01**	**0.000**	STC_OLS	**1.50**	**0.000**	STC_OLS	**0.53**	**0.000**
RR	**0.69**	**0.001**	RR	**0.98**	**0.000**	RR	**0.36**	**0.000**
SAS_RR	**1.06**	**0.000**	SAS_RR	**1.27**	**0.000**	SAS_RR	**0.59**	**0.000**
STC_RR	**1.02**	**0.000**	STC_RR	**1.49**	**0.000**	STC_RR	**0.54**	**0.000**
SKTT	**0.66**	**0.000**	SKTT	**0.96**	**0.000**	SKTT	**0.32**	**0.000**
KTRL	**0.70**	**S**	KTRL	**0.93**	**S**	KTRL	**0.37**	**S**

Note: KTRLine does not report P-values, therefore if the 95% CI did not contain zero it was considered significant (S).



Discussion:

Matlacha Pass Aquatic Preserve:

There was good agreement among the various trend analyses for DO in the Matlacha Pass Aquatic Preserve; all of which showed a declining trend between -0.6 and -1.5% per year. This suggests that the significant increasing trend in summer temperatures (0.31°C per year) was driving down DO saturations. In addition, increasing trends in chlorophyll *a* (between 0.66 and 1.06 µg/L per year) may have caused conditions to trend toward hypoxia (Lowery 1998). A declining trend in DO concentrations (-0.3 mg/L per year) within Matlacha Pass was also noted for the Coastal Charlotte Harbor Monitoring Network (CCHMN) from 2001 through 2007 (Leary 2009). However, the overall average DO saturation (87%) suggests that the estuary was still healthy (Cary Institute of Ecosystem Studies 2009). In waterbodies, DO is produced primarily by dissolution from the atmosphere (Stickney 1984), through wind and wave action, and to a lesser extent by photosynthesis; and may serve as an indicator of biotic productivity and estuarine health (e.g., Stickney 1984 and Colling 2006). Super saturation of DO can occur due to high rates of photosynthesis and surface mixing caused by wind and wave activity (e.g., Garcia and Gordon 1992). In the Charlotte Harbor estuarine complex, low DO values may be due to the combined effects of high water temperature, high levels of colored dissolved organic matter (CDOM) and anthropogenic sources of nutrients and other materials (Morrison et al 1998, Camp, Dresser & McKee, Inc. 1998 and Leary 2009). Therefore, declines in DO should be investigated.

Temperature was multimodal and, as noted earlier, summer temperatures were significantly increasing, whereas winter temperatures were trending down; suggesting increasing environmental variability. This increasing variability in temperature may prove to be detrimental to certain organisms in the region (e.g., Roessig 2004, Folguera et al 2009, Florida Oceans and Coastal Council 2009, Nye 2010, Estay et al 2011, Folguera et al 2011). Temperature is one of the most important properties in water (e.g., Stickney 1984 and Colling 2006), and is a controlling factor in a many chemical and biological processes within estuaries and may serve as a biological predictor (Duffey et al 2007). For example, temperature plays a key role in seasonal primary productivity and triggers life stage changes in many finfish and shellfish species. In general, temperature varies with: depth, mixing due to wind and tides, season, time of day, and anthropogenic influences (EPA 1993). Further analyses of temperature with respect to individual seasonal trends are required.

There was no significant trend in salinity, and there was little agreement among the analyses. There was good agreement between all models and trend analyses for pH, indicating a decrease of between -0.02 and -0.04 SU per year. Leary (2009) noted a declining trend for pH within Matlacha Pass from 2001 – 2007 at a rate of -0.08 SU per year, as well. As with temperature, the decline in pH may also be indicative of climate change (since there was no significant trend in salinity) due to increased dissolved CO_2 thereby acidifying the water (e.g., Colling 2006 and Nye 2009). The pH of the waterbody affects concentrations of a variety of biological and chemical substances (Stickney 1984). In addition, pH can be influenced by the local geology and hydrology of the watershed (e.g., Stickney 1984 and Boggs 2006). Many aquatic organisms are adapted to narrow pH ranges, with optimal pH ranges for many fish being about 6.5-8.5 (e.g., Chamberlain et al 1980). Furthermore, changes in pH in aquatic systems are positively linked with changes in salinity and photosynthesis (Stickney 1984, Day et al 1989). Abnormal pH values have direct and indirect detrimental effects upon the estuarine ecosystem and changes in pH should be investigated (Day et al 1989).

Turbidity within Matlacha Pass Aquatic Preserve appears to be decreasing, however assumptions were typically violated and nonparametric tests were not significant. Excessive turbidity can have a variety of effects on physical and biological processes, including: increased light attenuation (Kirk 1983, McPherson and Miller 1994, Ott et al 2006), and potentially decreasing the productivity, growth and survival of shellfish (Volety and Encomio 2006). Freshwater inflows from rivers are primary sources of sediment loading into the Charlotte Harbor estuarine complex (Hammett 1990), with nonpoint sources contributing almost 97% of the total suspended solids (Squires et al 1998). However, resuspension of sediments due to wind and wave action may be the primary cause for increased turbidity in shallow estuaries, such as Matlacha Pass (de Jorge and Beusekom 1995). Turbidity results should continue to be monitored and compared with other programs within the Matlacha Pass Aquatic Preserve.

There were significant declining trends for TN, TKN, and TP (-0.03, -0.03, and -0.005 mg/L per year, respectively) within the region, suggesting a reduction in nutrient loading to the region. The TP trend was the same rate and direction as found in 40 year analyses from 1970 to 2009 (Leary 2010). Furthermore, Leary (2009) documented a declining trend in TN levels (less than -0.001 mg/L per year) for the CCHMN from 2001 – 2007. In addition, for these three parameters there was good agreement between all the models and trend analyses. However, there was little agreement among the models and trend tests for NO_x and ammonia, with results trending in both directions. This may be due to the large number of results reported below the MDL and how the results were treated for analyses (Helsel 2010). Excess nitrogen could have serious impacts on Matlacha Pass Aquatic Preserve, primarily due to increased phytoplankton production, which may result in decreased water clarity affecting seagrasses (e.g., Jaworksi 1981, Montgomery et al 1991, Corbett 2005 and Doering et al 2006). McPherson and Miller (1990) noted that nitrogen may be influenced by salinity concentrations as well, with observed decreases in nitrite and nitrate at salinities greater than 20 PSU. In the Charlotte Harbor region, freshwater flow from major rivers is the major source for phosphorus entering the estuary, with the Peace River watershed contributing about 85% of total phosphorus loads into the Charlotte Harbor estuarine complex (Hammett 1990 and Morrison et al 1998). However, with declining trends in nutrients, other factors effecting water quality should take precedence, while continuing to monitor nutrients.

As stated earlier, there were significant increasing trends for chlorophyll *a*. This was true of all models and analyses. This result seems to contradict the results for the nutrients, however when the phytoplankton taken in nutrients, they remove it from the water column and are transport to other areas by the currents. Past studies within the Charlotte Harbor estuarine complex indicate that increased chlorophyll *a* concentrations are primarily due to increased inorganic nitrogen and vary in response to availability light within the water column (Montgomery et al. 1991, McPherson et al 1996, Doering et. al. 2006). Effects of increased chlorophyll *a* levels include: increased light attenuation due to increased phytoplankton within the water column (Doering et al 2006, Ott et. al. 2006); and hypoxia due to increased utilization of DO for decomposition of phytoplankton and increased community respiration (Doering et al 2006). Changes in water clarity can be attributed to fluctuations in CDOM, phytoplankton, non-algal suspended matter and fluvial inputs (Kirk 1983, McPherson and Miller 1987, McPherson and Miller 1994, Dixon and Kirkpatrick 1999, Doering and Chamberlain 1999, Ott et. al. 2006, and Duffey et al. 2007). The increasing trend in chlorophyll *a* must be tracked and controlled in order to ensure the health of the Matlacha Pass Aquatic Preserve.

MP1A

For the field parameters, there was little agreement between models and trend tests, nor were there any significant results for parametric or robust statistics when assumptions were met or for nonparametric statistics. This suggests that these physical parameters at MP1A were fairly stable. However, there was good agreement among all models and trend tests for TN, TKN, ammonia and TP. All of which indicated significant decreasing trends for these nutrients. This was not the case for NO_x, which had results trending both ways and none were statistically significant. Lastly, all models and analyses showed significant increases in chlorophyll *a*. As noted for the aquatic preserve, this may be due to the phytoplankton removing nutrients from the water column at other locations, and then drifting to the study site. This site is located approximately halfway between two seagrass transects, MP01 and MP02. Leary (2011a) documented that site MP01 was the most improved of the seagrass transects within the Matlacha Pass Aquatic Preserve, and that MP02 was the most degraded from 1999 – 2009; results from both studies should be examined in order to aid in management decisions for the aquatic preserve.

MP2B

There was a declining trend in DO at MP2B. This trend was true for most modeled results and all trend tests. In addition, all models and most trend analyses showed a decreasing trend in pH. Results for salinity, temperature and turbidity were inconclusive due to violations of assumptions and low statistical significance; however many of the models and trend test were in agreement and suggest that temperature and turbidity are decreasing while salinity is increasing. This may be due to a reduction in releases from the S-79 Franklin locks and dam to the tidal portion of the Caloosahatchee River (e.g., Brown et al 2012). There was a decreasing trend in TN and TKN, with all trend tests and modeled results in agreement. However, there was little agreement among the models and trend tests for NO_x and ammonia, with results trending in both directions and may be due to the large number of results reported below the MDL and how the results were treated for analyses (Helsel 2010). All models and trend tests showed declines in TP, but were either not statistically significant or they violated assumptions. And, as with the aquatic preserve as a whole and MP1A, there was a significant increase in chlorophyll *a*, and all results from all analyses indicated this same trend. Results from this site need to be compared with results from the nearby seagrass transect (MP04; approximately 0.6 km south of MP2B), which had significant increases in epiphyte densities and significant decreases in blade lengths from 1999 through 2009 (Leary 2011a).

MP3C

Although no trend analyses could be run on MP3C, comparisons were made between this site and the other two locations. Site MP3C had statistically significantly lower values than both MP1A and MP2B for the following parameters: DO, pH, TN, TKN, and TP. This site was not different from the other sites with regards to chlorophyll *a*, turbidity, salinity, and temperature. For both ammonia and NO_x MP3C was statistically the same MP1A but MP2B was significantly higher than the other two sites for ammonia and MP3c was significantly lower than MP2B for NO_x. From the results, it appears that there is very little flushing of this site, possibly due to the location of the tidal node in Matlacha Pass, as the other two locations are more similar to each other than to MP3C located between them. The low pH and DO results need to be examined with regards to MP02 (a seagrass transect located 1.6 km northwest of MP3C) which had significant declining abundances and blade lengths and increasing epiphyte densities from 1999 to 2009 (Leary 2011a).

Conclusions:

Declining trends for both pH and DO within the Matlacha Pass Aquatic Preserve and at site MP2B are of concern. However, these results need to be compared with rainfall during this time frame, as this can impact both parameters. In addition, these trends in DO and pH had been documented from an earlier study. Furthermore, significantly lower DO and pH results from site MP3C make these two parameters of specific interest, in particular how they relate to increased epiphyte densities and decreased blade lengths at nearby seagrass transects. Likewise, increasing chlorophyll *a* concentrations at MP1A, MP2B, and within the aquatic preserve as a whole require further attention and comparison with seagrass transect results. Temperature was found to be multimodal, and was increasing during summer months while decreasing during winter months. This means that there was increasing variability in temperature, and may stress organisms within the aquatic preserve. The lack of trends in salinity and turbidity suggest that these two parameters were stable during the study period. Whereas, declining trends in nutrients at both sites and within the Matlacha Pass Aquatic Preserve suggest improving water quality for the region. The same trends in nutrients have been documented in previous studies for the aquatic preserve.

Results for temperature, DO and pH may be indicative of climate change, and need further examination, as does chlorophyll *a*. Reductions in nutrients are most likely due to new fertilizer ordinances; best management practices with regards to agriculture; and continued education and outreach programs related to proper fertilizer use for homeowners, as well as improved stormwater retention. Comparisons between this program and other monitoring programs in the area need to be examined, and site specific trend analyses should be conducted for the CHEVWQMN. Further research and continued protection of the waters and submerged resources are required in order to maintain the socio-economic and ecological value of the Matlacha Pass Aquatic Preserve.

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