University of Puerto Rico
College of Agricultural Sciences
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DETERMINATION OF NUMERIC NUTRIENT TARGET CRITERIA IN LAKES AND RESERVOIRS OF PUERTO RICO

FINAL PROGRESS REPORT
ENCOMPASSING PERIOD FROM
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EXECUTIVE SUMMARY

The list of Impaired Waters of Puerto Rico (305(b)/303(d) states that all of our reservoirs fail to meet existing aquatic life criteria for dissolved oxygen. Eutrophication, a condition characterized by an elevated nutritional status and ensuing high productivity of the reservoirs was identified as the reason for impairment. However, numerical water quality standards establishing critical nutrient levels requiring remedial action are not available for Puerto Rico. The objectives of this project were to develop reference conditions for nutrients (nitrogen and phosphorus) in reservoirs of Puerto Rico, and evaluate the phytoplankton diversity in lakes of Puerto Rico linkages with nutrients.

A monitoring study was conducted using the complete reservoir (19) monitoring network of Puerto Rico. Several sections within each reservoir were sampled four times per year during the two years duration of this project. Samples were analyzed for several parameters including: Total Kjeldahl Nitrogen, Total Phosphorus, and Chlorophyll *a*. In accordance with EPAs National Nutrient Criteria program protocol the lower 25th percentile value of the frequency distribution of all of the lake data available for each parameter (i.e. TN, TP, SD, chlorophyll *a*) was selected as the reference condition.

The reservoir population of Puerto Rico was widely distributed in terms of their nutritional status. A preliminary ranking of the trophic status of the reservoirs was performed based to the trophic state index approach (Carlson, 1977). According on the TSI(TP) index the following reservoirs fell in the mesotrophic category: Carite, Cerrillo, Garzas, Guajataca, Matrullas, and Patillas. Reservoirs, Caonillas, Cidra, Curias, Guayabal, Guayo, Guineo, La Plata, Loco, Luchetti, Melania, Dos Bocas, and Toa Vaca ranked in the eutrophic group, whereas Carraizo ranked in the hypereutrophic category.

A significant difference between the concentration of nutrients at the riverine (entrance) and the lacustrine (near dam) sections of the reservoirs was observed with higher nutrient and chlorophyll *a* levels were observed at the entrance. The relationship between chlorophyll *a* values and nutrients was also stronger at this section (riverine). Estimates of reference conditions for TP, TKN, TN, and Chlorophyll *a* were 17.0 µg/L, 0.26 mg/L, 0.36 mg/L, and 2.87 µg/L, respectively, similar to values proposed by EPA for other ecoregions of the U.S.A. The large difference between the proposed value for phosphorus (17.0 µg/L) and the current water quality standard (1,000 µg/L TP) is noticeable. The adoption of numeric nutrient criteria will enable PREQB to identify nutrient impaired waters and prioritize remediate actions to restore the integrity of the waters and prevent further deterioration.

A series of *in-situ* nutrient response studies were conducted at Lago Guajataca to identify the limiting nutrient for phytoplankton growth on our reservoirs. Both nitrogen and phosphorus were shown to be limiting factors to aquatic biomass growth. The result contrasts with observations from temperate lakes, where phosphorus has been identified as the sole controlling factor to algae growth. A highly significant correlation among nutrients, as well as between nutrients and lake productivity (chlorophyll *a*) was observed during the monitoring period. A relationship between the concentration of total phosphorus (TP) in the reservoir vs. the total nitrogen (TN)/TP ratio resulted in two populations defined by a change point (17.1 µg/L) that is remarkably similar to the reference value proposed for TP (17.0 µg/L). Results demonstrate the importance of controlling loadings of both nutrients (N and P) into lakes of Puerto Rico.

A total of 70 taxa can be considered regular planktonic organisms in lakes of Puerto Rico. Of the 70 taxa identified, species richness was highest in the Chlorophyta (green algae) (34 taxa),

which mainly comprised taxa of the orders Desmidiales and Chrorococcales. Bacillariophyta had 16 taxa, Dinophyta had 7 taxa, Cyanobacteria had 7 taxa, and Chrysophyta were less diversified.

Microcystis was the only algal group found in all reservoirs. *Microcystis* are known to form algae blooms in eutrophic waters. Since some of the species are highly toxic preventive actions must be implemented to prevent the formation of blooms by these algae. Other genera that were observed in high frequency in most lakes were the genera *Fragilaria*, *Gomphonema* and *Pediastrum*, which are indicators of eutrophic conditions, and usually appear under high N/P ratios.

In terms of abundance, the dominant species were those belonging to the genera *Pediastrum, Peridinium* and *Botryococccus.* Overall, the mean relative proportional composition (%) was as follows: Chlorophyta > Dinophyta > Ochrophytes > Cyanobacteria > Euglenophyta. The division Chlorophyta had the higher representation in all samplings. The fact that Chlorophytas were found in highest numbers, and that the types of alga found within Chlorophyta division are associated with eutrophic conditions, suggests that there is a high probability for present and potential water-quality degradation of our reservoirs.

A relationship between the mean TP concentrations of the reservoirs versus their TKN/TP ratios, resulted in a negative exponential relationship. The lakes associated with eutrophy and hypereutrophy, namely Lago Carraizo, Dos Bocas and La Plata clearly had lower TKN/TP ratios, whereas the lakes associated with hypotrophy and mesotrophy had greater TKN/TP ratios and lower TP concentrations. The results suggest a N limiting condition in eutrophic and hypereutrophic lakes and a P limiting condition in hypotrophic and some mesotrophic lakes.

A significant difference in algal community structure was observed in reservoirs with low TKN/TP ratios (i.e., Lago Carraizo, Dos Bocas, La Plata and Caonilla) in comparison to reservoirs with high TKN/TP ratios (Carite, Guajataca, and Patillas).

1.0 GENERAL INTRODUCTION

According to the most recent list of Impaired Waters for Puerto Rico all reservoirs in the island are impaired for aquatic life use (PREQB 2003). The PREQB has identified low dissolved oxygen as the major cause of impairment. The report identifies nutrients as one of the primary causative agents of the eutrophication that lead up to the observed dissolved oxygen values, yet quantitative information is not included to support this. Interestingly, nutrients have yet to be officially identified as a major cause of impairment in Puerto Rico. Practically no water body is listed as being impaired by nutrients. This contrasts with estimates by the National Water Quality Inventory, which lists nutrients as the leading cause of water pollution in the continental United States. More than 3.4 million acres of lakes and reservoirs and 84,000 miles of rivers and streams are believed to be impaired as a result of excess nutrients at the national level. The fact that nutrients are not regarded as a major cause of pollution in Puerto Rico may be due to the lack of adequate standards that allow for the identification of nutrient impaired waters. The current water quality standard for phosphorus in Puerto Rico, 1000 µg total P L^{-1}, corresponds to the maximum discharge load allowed to point sources by USEPA. In contrast, numeric phosphorus criteria adopted in other regions of the U.S. place this value between 8 and 40 µg P L^{-1} for lakes (USEPA, 2000).

Although phosphorus is usually the limiting factor for aquatic biomass growth in surface waters, nitrogen is also a major controlling factor. In Puerto Rico, a 10 mg L^{-1} NO_3 is used as drinking water criteria. However, numeric criteria aimed at preserving the “natural” status and protecting the biological and chemical integrity of surface waters have yet to be established. Numeric criteria proposed for other ecoregions of the U.S. range between 0.1 – 1.68 mg/L TN for lakes, and between 0.12 to 2.18 mg/L for rivers (USEPA, 2000), values well below our current standard.

In 1994, the U. S. Environmental Protection Agency (USEPA) established the National Nutrient Criteria Program. The goal of this program is to reduce cultural eutrophication by developing guidelines for the establishment of numeric nutrient criteria at a state (tribal) level. The criteria, which represent conditions of waters minimally impacted by human activities, will enable regulatory agencies to identify, prioritize and restore nutrient impaired waters. Because different regions in the U.S. differ greatly in terms of the parameters that define the limnology of a particular water body (e.g., soil type, parent material, precipitation regimes, water body morphometry), USEPA adopted a regional approach to develop their nutrient criteria. Initially, the continental U.S. was divided into 14 separate ecoregions of similar geological and geographical characteristics. Hawaii, Alaska and U.S. Trust Territories (including Puerto Rico) were to be covered under separate ecoregions. States and territories of the U.S. should use the numeric criteria developed by USEPA as a basis for the development of site-specific criteria to protect the designated uses of those waters. Once developed, these criteria should become part of the State Water Quality Standards. Several ecoregions have already published their numeric criteria (www.epa.gov). However, at the time this project started USPEPA had yet to initiate the process for establishing the nutrient criteria for Puerto Rico.

The objective of the study was to establish a reference condition that could serve as framework for the development of numeric criteria for nutrients (nitrogen and phosphorus) for lakes/reservoirs of Puerto Rico.

1.1. OBJECTIVES

1. Develop reference conditions for nutrients (nitrogen and phosphorus) in lakes/reservoirs of Puerto Rico.
2. Evaluate the phytoplankton diversity in lakes of Puerto Rico and establish linkages with nutrients.

1.2. EXPERIMENTAL APPROACH

In accordance with USPEA Numeric Criteria Guidelines a monitoring study was conducted using the complete reservoir (19) monitoring network of Puerto Rico (Table 1.1). At least two monitoring stations within each lake (entrance and dam) were established in most reservoirs. The biggest reservoirs (> 100 ha) usually have three stations, corresponding to the riverine (entrance), transitional (center), and lacustrine (near dam) sections of the lake. Few of the smallest reservoirs (surface area < 30 ha) have only one monitoring station. Each station was monitored four times per year for the two years duration of this project. The goal was to sample during "rain", "dry" and transitional periods during the year. Samples were collected at the surface (≤ 1m) by PREQB personnel according to the procedures described in PREQB SOP No. 023. Analyses for pH, dissolved oxygen and conductivity were conducted at the sampling site according to PREQB Field Measurement SOPs 021.1, 021.2, 021.3 and 021.4 respectively.

After adding the specified preservatives all samples were placed in an ice chest containing sufficient ice to cool the samples to 4°C and transported to the PREQB Lab. Samples remained under PREQB custody until transferal to authorized AES lab personnel under appropriate chain of custody procedure.

Samples were analyzed for: total Kjeldahl nitrogen (TKN)(EPA method 351.2), total phosphorous (TP)(EPA method 365.4), and chlorophyll "a" (Chl *a*)(EPA method 445.0). Estimates of the amount of dissolved organic carbon (DOC) were obtained by quantifying the absorbance (280nm) of filtrated samples (<0.45μm cellulose fiber filter) on a DU-520 Beckman UV/VIS spectrophotometer after calibration with an organic carbon standard (Chin et al., 1994). Specific details on analysis methodology are included in the Quality Assurance Project Plan of this study. In addition, an evaluation of the phytoplankton diversity in the lakes was conducted (Chapters 3 and 4).

The reference condition was established according to the approach recommended by the National Nutrient Criteria program (USEPA, 2000). The lower 25th percentile value of the frequency distribution of all of the lake data available for each parameter (i.e. TN, TP, SD, chlorophyll *a*) was selected as the reference condition. The reference conditions proposed herein are one component of the numeric criteria establishment process. The criteria development process may include up to five separate components, namely:

- Investigation of the historical record
- Establishment of reference condition
- Use of models – (recommended when existing data is unavailable or when an adequate representation of the population of lakes/reservoirs is lacking)
- Expert Assessment of Information
- Assessment of downstream effects

Table 1.1. PREQB reservoir monitoring network.

Lake No.	Lake	Station Number	Description
1	Luchetti	89017 89018 89019	Near Río Yauco Center Dam
2	Loco	89020 89021	Near Quebrada Grande Near Dam
3	Patillas	89022 89023 89024 89025	Near Río Patillas Center Near Dam Near Río Marín
4	Las Curías	89027 89028	East side at the end of lake Near dam, near San Juan
5	Cidra	89029 89030 89031	Near the second bridge Center Near Dam
6	Caonillas	89001 89002 89003	Near Río Caonillas Center Dam
7	Guayo	89004 89005 89006	Near Río Guayo, near Adjuntas Center Dam
8	Guineo	89007 89008	Near Río Toro Near Dam
9	Matrullas	89009 89010	Near Río Matrullas Near Dam
10	Guayabal	89011 89012 89013	Near Río Jacaguas Center Dam
11	Cerrillos	89032 89033 89034	Near Río Cerrillos Center Dam
12	Toa Vaca	89014 89015 89016	Near Río Toa Vaca Center Dam
13	Guajataca	50010720 50010790	Near Río Dam
14	Garzas	50020050	Dam
15	Dos Bocas	50025110 50027090	Near Río Grande the Arecibo Near Dam
16	La Plata	50044400 50044950	Near Río la Plata Near Dam
17	Carite	50039900 50039950	Near Río la Plata Near Dam
18	Loíza	50057500 50058800	Near Río Center
19	Melanía	89026	Center

2.0 CHEMICAL COMPOSITION OF LAKES OF PUERTO RICO

2.1. Sampling dates

A total of eight sampling events were conducted on this project. Results have been included in previous progress reports (Table 2.1). The complete data set is presented in Appendix A.

Table 2.1. Sampling events completed to this point:

Sampling event	Encompassing dates	Progress reports submitted
1	8/12/03 – 9/2/03	
2	11/7/03 – 12/10/03	11/25/03
3	2/23/04 – 3/18/04	5/29/04
4	6/7/04 – 6/21/04	
5	8/9/04 – 8/25/04	
6	11/16/04 – 12/9/04	12/27/2004
7	2/25/05 – 3/18/05	5/30/2005
8	6/7/05 – 6/17/05	

2.2. Descriptive Statistics

Descriptive statistics of water-quality data collected for all lakes are described in Table 2.2, and for individual lakes in Table 2.3.

Table 2.2. Statistical summary of chemical analyses results of lake samples.

Parameter	N	Median	Mean	25th percentile
Chlorophyll a (µg/L)	282	5.67	11.07	2.87
TKN (mg/L)	328	0.36	0.39	0.26
TP (mg/L)	328	0.028	0.038	0.017
DP (mg/L)	326	0.01	0.013	0.005
DOC (mg/L)	328	2.72	3.38	2.11
TSI (Chlorophyll a)	282	47.62	48.12	40.96
TSI (TP)	328	52.07	52.58	45.01

Table 2.3. Statistical summary of chemical analyses for individual lakes.

Lake	**Parameter**	**Chloro-phyll a (µg/L)**	**TKN (mg/L)**	**TP (mg/L)**	**DOC (mg/L)**	**TSI (Chl *a*)**	**TSI (TP)**
Curias	Mean	8.04	0.50	0.030	5.79	43.06	51.81
	Median	3.31	0.44	0.028	5.50	42.32	52.20
	25th per. [1]	2.35	0.35	0.020	4.46	38.98	47.28
La Plata	Mean	15.19	0.39	0.051	5.50	51.51	58.95
	Median	6.29	0.35	0.058	3.84	48.30	62.70
	25th per.	3.32	0.28	0.030	3.34	42.37	53.20
Carraízo	Mean	14.21	0.62	0.122	5.35	50.53	71.31
	Median	6.73	0.51	0.121	5.35	49.30	73.19
	25th per.	5.76	0.33	0.085	4.65	47.78	68.02
Carite	Mean	2.85	0.29	0.013	2.90	38.72	40.60
	Median	2.27	0.26	0.013	2.60	38.60	41.09
	25th per.	1.22	0.23	0.011	2.14	32.54	38.04
Patillas	Mean	2.81	0.26	0.017	2.18	38.62	44.00
	Median	2.31	0.27	0.016	2.07	38.81	44.13
	25th per.	1.63	0.19	0.011	1.78	35.41	38.67
Guayo	Mean	13.32	0.41	0.032	3.04	51.01	52.14
	Median	8.41	0.42	0.030	2.42	51.29	53.20
	25th per.	4.56	0.27	0.019	2.14	45.49	46.61
Caonillas	Mean	14.71	0.42	0.04	3.05	53.48	55.12
	Median	10.51	0.42	0.04	2.66	53.68	57.34
	25th per.	6.13	0.31	0.021	2.27	48.39	48.05
Matrullas	Mean	9.26	0.32	0.026	2.00	47.99	49.75
	Median	6.49	0.34	0.024	1.82	48.89	50.13
	25th per.	3.11	0.25	0.018	1.61	41.73	45.81
Cerrillo	Mean	7.81	0.36	0.026	2.18	44.83	49.25
	Median	4.13	0.31	0.022	1.97	44.50	48.72
	25th per.	2.60	0.26	0.016	1.84	39.97	44.13
Cidra	Mean	11.07	0.49	0.040	5.62	48.10	55.18
	Median	7.01	0.43	0.035	5.18	49.71	55.42
	25th per.	3.05	0.40	0.024	4.37	41.53	49.67
Guineo	Mean	23.79	0.46	0.034	3.65	55.17	53.32
	Median	9.58	0.41	0.030	3.08	52.74	53.06
	25th per.	4.74	0.24	0.022	2.73	45.87	48.71
Guajataca	Mean	4.27	0.29	0.017	2.53	42.45	43.90
	Median	3.35	0.29	0.015	2.41	42.45	43.20
	25th per.	2.35	0.25	0.013	2.14	38.99	40.57
Dos Bocas	Mean	15.54	0.43	0.070	2.87	51.46	60.83
	Median	7.01	0.36	0.050	2.07	49.70	60.55
	25th per.	5.57	0.23	0.028	1.45	47.44	51.82

Table 2.3 (cont.).

Lake	**Parameter**	**Chlorophyll a (µg/L)**	**TKN (mg/L)**	**TP (mg/L)**	**DOC (mg/L)**	**TSI (Chl *a*)**	**TSI (TP)**
Melania	Mean	4.33	0.41	0.028	3.95	44.52	50.32
	Median	4.76	0.45	0.025	3.75	45.90	50.09
	25th per.	2.87	0.27	0.018	3.10	40.96	45.81
Loco	Mean	8.13	0.40	0.045	2.97	49.74	55.36
	Median	7.90	0.30	0.036	2.67	50.88	55.82
	25th per.	4.36	0.27	0.027	1.76	45.04	51.68
Luchetti	Mean	22.23	0.42	0.044	2.89	59.19	55.53
	Median	19.68	0.47	0.032	2.25	59.83	54.23
	25th per.	10.48	0.25	0.026	2.00	53.65	50.85
Garzas	Mean	6.98	0.26	0.021	1.95	47.24	47.10
	Median	5.94	0.27	0.018	1.81	48.08	45.62
	25th per.	4.25	0.17	0.015	1.39	44.78	43.19
Guayabal	Mean	15.07	0.33	0.033	2.63	50.58	51.50
	Median	11.60	0.27	0.029	2.62	53.63	52.71
	25th per.	3.22	0.08	0.01	1.76	40.53	37.35
Toa Vaca	Mean	6.07	0.39	0.043	3.40	44.35	55.03
	Median	3.79	0.39	0.041	3.20	43.66	57.70
	25th per.	2.65	0.30	0.024	2.90	40.15	49.79

[1] Indicates 25th percentile of the data.

A statistical analysis was performed to identify within lake, among lake, among sampling dates, or sampling season differences, as well as to evaluate correlations between different chemical, biological and/or physical parameters. A non-parametric test with paired data (Wilcoxon Signed Rank) was used to establish differences between rivering (entrance) and lacustrine (dam), and transitional (center) and (lacustrine) since ANOVA residuals were not uniform. A summary of that analysis will be discussed next (the complete statistical report is shown in Appendix B).

Significant differences (95%) between the riverine and lacustrine areas of lakes were observed for all the parameters (Secchi depth, TP, TKN, DP, DOC, N/P ratio, pH and TSITP), except electrical conductivity and dissolved oxygen (DO) (Table 2.4). Differences between the transitional and lacuscrine sections were less evident, with only TP, DOC, and TSITP denoting significant differences. The results are in accord with the general behavior observed in most lakes, with higher nutrient concentrations at the lake entrance than at other reservoir sections. This generally results in chlorophyll *a* values being higher, and dissolved oxygen and Secchi depth values being lower at the entrance than close to the dam.

Table 2.4. Wilcoxon Signed Rank mean values and significance levels between riverine and lacustrine lake areas. P-value < 0.05 indicate significant differences between stations.

Parameter	**Mean value riverine**	**Mean value lacustrine**	**P-value**
E.C. µmhos/cm	201.00	178.50	0.25
pH	7.97	8.13	3.34E-3
Secchi Depth (m)	1.02	1.53	4.52E-15
DO (mg/L)	7.45	7.53	0.21
TP (mg/L)	0.048	0.029	3.48E-16
TKN (mg/L)	0.43	0.36	1.17E-6
DP (mg/L)	0.019	0.008	1.18E-4
DOC (mg/L)	3.73	3.12	1.33E-8
Chl a (µg/L)	13.78	8.91	2.03E-3
TKN/P	12.43	15.37	2.49E-5
TSI Chl a	49.88	46.66	1.88E-3
TSI TP	55.83	49.50	4.72E-17

2.3. Relationships among water-quality parameters

Spearman correlation coefficients for the complete data set show a weak but positive correlation ($r = 0.63$) between TP and Chl *a* (Table 2.5) (see Appendix B for complete statistical results). This indicates that as total P loadings into the lakes increase, an increase in algae biomass density and overall lake productivity will result. As expected, increases in algae biomass density (as measured by Chl *a*) diminish the light penetration depth into the lake profile (SD). This is evidenced by the negative correlation coefficient obtained between these parameters. In fact, both Chl *a* and TP are negatively correlated with SD.

Table 2.5. Spearman correlation coefficients between *specific* descriptive parameters of lake trophic status; **all sampling stations combined**.

	TP	**TKN**	**SD**
Chl *a*	0.63	0.37	-0.32
TP	1.00	0.56	-0.40

A separate correlation analyses was performed according to lake location to evaluate the relationship between the different parameters. Results for station 1 (lake entrance) were similar to those obtained with the complete data set (Table 2.6). A positive correlation between TP and Chl *a*, and a negative correlation between both parameters (TP and Chl *a*) and SD was observed. In addition there was a positive correlation between TP and TKN, suggesting that similar transport factors are responsible for a significant portion of the nutrient loadings into the lakes.

Table 2.6. Spearman correlation coefficients between *specific* descriptive parameters of lake trophic status; **data from riverine station only**.

	TP	**TKN**	**SD**
Chl *a*	0.47	0.39	-0.29
TP	1.00	0.56	-0.45

Reductions in SD values may be caused either by an increase in algae biomass concentration or by inorganic (sediment) particles coming into the lake as a result of runoff events. High sediment loads will mask the effect of nutrients on aquatic biomass development, thus reducing the quantitative relationship between nutrients and biomass. To evaluate the potential interference effect of sediment loads on our relationships a separate analysis was conducted excluding samples with SD less than 1. At this station (riverine) a significant improvement in the TP- Chl *a* relationship was observed when data with SD<1 were removed from the analysis (Table 2.7). This suggests that a significant fraction of the TP coming into the reservoirs is in particulate form and is not immediately available for algae use. Such mechanism should be more evident at the entrance, and, as the results indicate, it looses relevance away from the entrance (center and lacustrine zones) as sediment particles settle into the reservoir bottoms (Tables 2.8 to 2.11). Removing the SD<1 data from the analysis weakened the relationships between TP and Chl *a* with SD. This suggests that, in general, light penetration at this station (entrance) is more significantly influenced by algae biomass than by suspended sediment particles.

Relationships between chemical and biological parameters at the center and lacustrine sections of the reservoirs were comparable (Tables 2.8, and 2.10). In this case however removing SD<1 data from the analyses weakened the relationships between TP, and TKN (Tables 2.9, and 2.11). As indicated previously this is a result of the effect of suspended sediment particles loosing their relevance in the deeper portions of the reservoirs.

Table 2.7. Spearman correlation coefficients between *specific* descriptive parameters of lake trophic status; **data from riverine station with Secchi depths values ≥ 1m**.

	TP	**TKN**	**SD**
Chl *a*	0.68	0.54	-0.06
TP	1.00	0.52	-0.18

Table 2.8. Spearman correlation coefficients between *specific* descriptive parameters of lake trophic status; **data from transition (center) section**.

	TP	**TKN**	**SD**
Chl *a*	0.48	0.23	-0.33
TP	1.00	0.52	-0.34

Table 2.9. Spearman correlation coefficients between *specific* descriptive parameters of lake trophic status; **data from transition (center) with Secchi depths values ≥ 1m**.

	TP	**TKN**	**SD**
Chl *a*	0.46	0.37	-0.19
TP	1.00	0.31	-0.10

Table 2.10. Spearman correlation coefficients between *specific* descriptive parameters of lake trophic status; **data from the lacustrine section only**.

	TP	**TKN**	**SD**
Chl *a*	0.32	0.21	-0.25
TP	1.00	0.53	-0.31

Table 2.11: Spearman correlation coefficients between *specific* descriptive parameters of lake trophic status; **data from the lacustrine section with Secchi depths values ≥ 1m**.

	TP	**TKN**	**SD**
Chl *a*	0.32	0.39	-0.16
TP	1.00	0.51	-0.11

2.4. Determination of Reference Conditions

Table 2.12 shows estimates of the reference conditions as determined by the lower 25th percentile value of the frequency distribution of all of the lake data available (all sampling stations (riverine, center, and lacustrine zone) and sampling events for each parameter. Predicted values are close to reference values proposed in other regions of the USA. In the case of nitrogen the value included is for Total Kjedahl Nitrogen (TKN), rather than for Total Nitrogen (TN) as in other ecoregions. An estimate of the reference condition for TN can be obtained by adding the concentration of nitrate to the TKN values of the samples and evaluating the distribution of the population of values as in the other cases. Our project did not contemplate analyzing the samples for nitrate. Rather we expected to use results from analyses performed by the PREQB laboratory on duplicate samples. However at the time of writing this report we had not received a PREQB-lab. report on nitrate analyses. Another potential source of information for establishing the TN reference condition was the PREQB reservoir historical data base. However, a quality assurance evaluation conducted by this research team on the data revealed a series of inconsistencies that were considered significant enough to defer use of the data for evaluation purposes. As a precautionary measure we did sent samples from 7 of the eight sampling events for nitrate analyses to external laboratories. Results of average nitrate concentrations for the different lakes are shown in Table 1.12 (raw data is included in Appendix A). Also included are results from average values of historical data compiled by USGS www.usgs.gov. In general, results from USGS are comparable to results obtained in this study.

Table 2.12. Reference condition values for lakes/reservoirs of Puerto Rico. Values for eight of the 14 USEPA ecoregions[1] are included for comparative purposes (refer to www.epa.gov for details on the ecoregions report).

Item	**PR[1]**	Eco II	Eco VI	Eco VII	Eco VIII	Eco IX	Eco XI	Eco XII	Eco XIII
TP ug/l	**17.0**	8.75	37.5	14.75	8.0	20.0	8.0	10.0	17.5
TN mg/l	**0.26[2]**	0.10	1.68	0.66	0.24	0.36	0.46	0.52	1.27
Chl *a* ug/l	**2.87**	1.90	8.59	5.23	2.39	5.18	2.79	2.60	3.35

[1] – ER II (Western Forested Mountains); ER VI (Corn Belt and Northern Great Plains; ER VII (Mostly Glaciated Dairy Region); ER VIII (Nutrient Poor Largely Glaciated Upper Midwest and Northeast); ER IX (Southeastern Temperate Forested Plains and Hills); ER XI (Central and Eastern Forested Uplands); ER XII (Southern Coastal Plain); ER XIII (Southern Florida Coastal Plain).

[2] – Value for Total Kjedahl Nitrogen (TKN)

Table 2.13. Average nitrate concentrations for lakes/reservoirs of Puerto Rico (analyses performed by an external laboratory.

Reservoir	**Average Nitrate Concentration (mg/L)**	**Average of Historical data. Source USGS**
La Curias	0.087	NA
La Plata	0.21	≤0.33
Carraízo	0.60	≤0.48[1]
Carite	0.11	≤0.10[1]
Patillas	0.07	NA
Guayo	0.24	NA
Caonillas	0.19	NA

Table 2.13 (cont.).

Reservoir	**Average Nitrate Concentration (mg/L)**	**Average of Historical data. Source USGS**
Matrullas	0.38	NA
Cerrillo	0.05	NA
Cidra	0.12	≤0.13[1]
Guineo	0.05	NA
Guajataca	0.10	< 0.10 (detection limit)[1]
Dos Bocas	0.44	≤0.33[1]
Melania	0.10	NA
Loco	0.62	NA
Luchetti	0.42	NA
Garzas	0.19	≤0.11[1]
Guayabal	0.18	NA
Toa Vaca	0.04	NA

[1] – Average values obtained by assigning the specified detection limit as the actual value for samples that fell below said limit. For instance, samples reported as < 0.1 were considered as 0.1 for calculation purposes.

Table 2.14 shows the reference condition obtained for TN when considering the nitrate results. Again results fall within the range of values reported for similar ecoregions (IX, XII, and XIII) in the U.S.A. In our perception the use of both reference conditions (TKN, and TN) for nitrogen could be a more effective tool for detecting nutrient impaired reservoirs in Puerto Rico, as nitrate concentrations does not seem to be significant in all reservoirs. In fact, lakes exhibiting the highest nitrate average concentrations (Carraizo, Luchetti, Loco, Dos Bocas, Matrullas) are on watersheds with extensive crop production or urban/suburban development. This suggests that in some of those lakes such concentrations may not be a reflection of a natural condition.

Table 2.14. Reference condition values for lakes/reservoirs of Puerto Rico. Values for eight of the 14 USEPA ecoregions[1] are included for comparative purposes (refer to www.epa.gov for details on the ecoregions report).

Item	**PR[1]**	Eco II	Eco VI	Eco VII	Eco VIII	Eco IX	Eco XI	Eco XII	Eco XIII
TP ug/l	**17.0**	8.75	37.5	14.75	8.0	20.0	8.0	10.0	17.5
TN mg/l	**0.36**	0.10	1.68	0.66	0.24	0.36	0.46	0.52	1.27
Chl *a* ug/l	**2.87**	1.90	8.59	5.23	2.39	5.18	2.79	2.60	3.35

It is important to notice the large difference between the reference condition for phosphorus (17.0 µg/L) and the current water quality standard in the island (1,000 µg/L). This has definitely prevented the identification of nutrient impaired waters in the island.

Although a significant difference in nutrient concentrations was observed between the riverine (entrance) section of the lakes, and the other sections (center and lacustrine (dam)) estimates of reference conditions excluding the riverine data does not yield significantly different results (TP – 16.4 µg/l; TN – 0.34 mg/L; Chl a – 2.80 µg/l). Thus, it is recommended that a single set of reference conditions be used.

A ranking of the different lakes based on TP, TKN, and Chl *a* concentrations, reveals that Carite, Patillas and Guajataca are the least nutrient impacted lakes; Carraízo, Dos Bocas and La Plata the more impacted lakes; and the rest of the lakes exhibit various degrees of impact from antropogenic activities (Table 2.15).

The Nutrient Criteria Program recommends establishing reference conditions for two independent variables (i.e., TP, and TN) and two response variables (e.g., Chl *a*, SD, DO, algae biomass, etc). Most ecoregions have chosen to use Chl a, and SD as their response variables. In our case the use of SD does not seem to be an appropriate alternative. Most reservoirs in Puerto Rico are subject to substantial sedimentation (Soller-López, 2002). The presence of significant amounts of inorganic particles interferes with light penetration and prevents the use of SD as a probe of phytoplankton productivity.

An alternative to assess compliance with aquatic life criteria on our reservoirs would be the use of dissolved oxygen (DO). Dissolved oxygen is a critical component to all aquatic life and is an essential indicator of aquatic ecosystem health. Hypoxia, commonly defined as water with DO concentrations <2mg/L (USEPA, 2003), is a symptom of a problem generally caused by excessive nutrient, and organic matter loadings into a water body. As a result, DO values have traditionally been used as an index of trophic status in temperate regions as the U.S.A. A cautionary note to using DO as an index of trophic status in the tropics has been raised by several researchers. For instance, Towsend indicated that trophic classifications based on hypolimnetic oxygen content were not universally applicable because of the temperature dependence of oxygen dynamics, and that hypolimnetic anoxia could not be used as an index of eutrophic conditions in the tropics (Townsend, 1996, 1999).

Tropical waters are more susceptible to oxygen depletion than their temperate counterparts, making the phenomena of lake/reservoir anoxia more prevalent in the tropics (Townsend, 1996). This is a result of the reduced solubility of oxygen in warm waters coupled with higher rates of microbial metabolism experienced at higher temperatures (Townsend, 1996, 1999). Those effects are compounded by long periods of stratification commonly experienced by some tropical water-bodies.

DO is still a very valuable tool to assess the toxicological impact on aquatic life, and should exclusively be used in that context in the tropics. The well documented substantial differences in DO dynamics between tropical lakes/reservoirs and temperate systems demand a thorough characterization of the DO dynamics in our reservoirs prior to adopting guidelines developed in temperate ecosystems.

Table 2.15. Ranking of lakes according to their chemical and biological properties. Numbers vary from the more impacted (1) to the least impacted lake (19) on each category.

LAKE	**Ranking for TP (mg/L)**	**Ranking for TKN (mg/L)**	**Ranking for Chl_a (µg/L)**	**Ranking for DOC (mg/L)**	**Ranking for TKN/TP**	**Ranking for TSIChl**	**Rank-ing for TSITP**
Caonillas	8	7	6	8	12	3	7
Carite	19	16	18	11	1	18	19
Carraizo	1	1	7	4	19	8	1
Cerrillo	15	13	13	17	6	13	15
Cidra	7	3	9	2	7	10	6
Curias	12	2	12	1	3	16	11
Dos Bocas	2	5	3	13	18	5	2
Garzas	16	18	14	19	10	12	16
Guajataca	18	17	17	15	2	17	18
Guayabal	10	14	5	14	16	7	12
Guayo	11	8	8	9	5	6	10
Guineo	9	4	1	6	11	2	9
La Plata	3	11	4	3	17	4	3
Loco	4	10	11	10	15	9	5
Luchetti	5	6	2	12	13	1	4
Matrullas	14	15	10	18	9	11	14
Melania	13	9	16	5	8	14	13
Patillas	17	19	19	16	4	19	17
Toa Vaca	6	12	15	7	14	15	8

A preliminary ranking of the lake trophic status was performed by means of the trophic state index approach (Carlson, 1977). The trophic state index uses phytoplankton biomass and P concentration as a basis for a continuum of trophic states of lakes and reservoirs. An inverse relationship between TP and transparency is assumed, with a doubling of TP for every half unit of secchi disk transparency. Chlorophyll pigments double every seven units. Trophic state index values for chlorophyll *a* and total P were calculated using the equations developed by Carlson for temperate regions lakes. The equations employed were:

$$TSI(Chl) = 10\left(6 - \frac{2.04 - 0.68 \ln Chl}{\ln 2}\right) \quad [1]$$

$$TSI(TP) = 10\left(6 - \frac{\ln \frac{48}{TP}}{\ln 2}\right) \quad [2]$$

A trophic state status is assigned based on the following categories (Table 2.16).

Table 2.16. Trophic state classification according to the trophic state index

Trophic State Category	Trophic State Index Value
Oligotrophic	< 30
Hypotrophic	30 – 40
Mesotrophic	40 – 50
Eutrophic	50 – 70
Hypertophic	> 70

Chlorophyll a and TP trophic state index values were calculated for each data point and the effects lake, lake location (entrance, center and damn areas), and season (dry or wet), on TSI(Chl) and TSI(TP) were evaluated. An analysis of the distribution of values between the different categories was conducted. Tables 2.17 and 2.18 show the category distribution for each lake based on the frequency values. Results from this exercise confirm the previous statement that most of our lakes would fall into the mesotrophic category with conditions that promote eutrophic behavior in particular zones of a significant number of lakes during different times of the year.

Table 2.17. Trophic state index for chlorophyll a, TSI(Chl), of lakes of Puerto Rico.

Trophic State Index Categories						
Lake name	**< 30**	**30 - 40**	**40 -50**	**50 – 60**	**60 - 70**	**>70**
Caonillas				X		
Carite		x				
Carraizo				X		
Cerrillo			X			
Cidra			X			
Curias			X			
Dos Bocas				X		
Garzas			X			
Guajataca			X			
Guayabal				X		
Guayo				X		
Guineo				X		
La Plata			X			
Loco				X		
Luchetti				X		
Matrulla			X			
Melania			X			
Patillas		x				
Toa Vaca			X			

Table 2.18. Trophic state index for total P, TSI(TP), values of lakes of Puerto Rico.

Trophic State Index Categories						
Lake name	**< 30**	**30 - 40**	**40 -50**	**50 – 60**	**60 - 70**	**>70**
Caonillas				x		
Carite			X			
Carraizo						x
Cerrillo			X			
Cidra				x		
Curias				x		
Dos Bocas					x	
Garzas			X			
Guajataca			X			
Guayabal				x		
Guayo				x		
Guineo				x		
La Plata				x		
Loco				x		
Luchetti				x		
Matrulla			X			
Melania				x		
Patillas			X			
Toa Vaca				x		

Overall, the trophic state index for TP was higher than for Chlorophyll. For TSI(Chl) as classified by lakes there were three main groups. In the hypotrophic group there was Patillas and Carite. In the mesotrophic group there were nine lakes, namely: Cerrillos, Cidra, Curias, Garzas, Guajataca, La Plata, Matrullas, Melania, and Toa Vaca. The rest of the reservoirs (i.e., Caonillas, Carraizo, Dos Bocas, Guayabal, Guayo, Guineo, Loco, and Luchetti) fell in the eutrophic group. The trophic state index by P was much more restrictive as all lakes fell in the mesotrophic group or higher. This reinforces what has been mentioned previously in that other factors, most possibly light, grazing, or other nutrients were limiting lake productivity in most of the reservoirs evaluated. This suggests that TSI(TP) may be a better index of potential lake productivity and consequently a better tool to judge nutrient impairment than TSI(Chl). The following reservoirs fell in the mesotrophic category based on TSI(TP): Carite, Cerrillo, Garzas, Guajataca, Matrullas, and Patillas. In the eutrophic group were the lakes: Caonillas, Cidra, Curias, Guayabal, Guayo, Guineo, La Plata, Loco, Luchetti, Melania, Dos Bocas, and Toa Vaca. Lago Carraizo was classified as hypereutrophic by this index.

There was a significant effect of location on the trophic state indices. The riverine portions of the lakes were significantly higher than the lacustrine lake areas with median TSI(Chl) values of 49 and 45, respectively, and with median TSI(TP) values of 56 and 49, respectively. Both TSI(Chl) and TSI(TP) values tended to be higher in wet than in dry conditions.

3.0. NUTRIENT DYNAMICS IN GUAJATACA LAKE

Guajataca is one of the few reservoirs in the island that can be considered to be minimally impacted by human activities. As results from this study attest nutrient levels at this reservoir remain near reference conditions. Based on that, we selected this reservoir decided to conduct an in-depth study on algae response to nutrients.

3.1 PERIPHYTOMETER STUDY

A series of *in-situ* assays were conducted to evaluate phytoplankton response to nutrients, and identify the limiting nutrient for phytoplankton growth on our lakes. Initially, two preliminary trials were conducted to evaluate the effect of different exposure times on chlorophyll *a* response using the Matlock periphytometer (Matlock et al., 1998, 1999). Three reactions times were evaluated, namely: 7 days, 10 days, 14 days. A detailed experimental protocol for these experiments follows:

The Matlock periphytometer floating rack was constructed using 7.62-cm (diameter) polyvinyl chloride (PVC) pipes and wire mesh (5 x 10 cm). The floating rack dimensions were 2.7 m (length) x 1.5 m (width) (Figures 3.1, and 3.2). The floatability of the rack was tested previously.

The experimental unit (periphytometer) consisted of a 1-L narrow-mouth bottle (Nalgene LDPE Narrow-Mouth, Fisher no. 02-9254-6F). A 2.54 cm diameter hole was drilled within the bottle, which permitted enclosing a 0.45 μm nylon membrane filter (Cole Palmer A-02916-44, which served as biofilter) and glass fiber filter (Whatman glass microfiber filter, Fisher 934 AH) as the growth substrate (Figure 3.3).

The floating rack contained 42 bottles in randomized complete block arrangement, with the bottles placed perpendicular to the expected current flow. The bottles were randomly assigned to one of two treatments which were blank (distilled water) and a nutrient solution (2 ppm of phosphorus and 14 ppm of nitrogen). The N solution was prepared with NH_4NO_3, and the P solution was prepared with NaH_2PO_4. Bottles from each of the two treatments/ replications were harvested at 7, 10, and 14 days after initiation of the experiment. The first trial was conducted on an irrigation pond at the Agricultural Experimental Station of the University of Puerto Rico at Isabela, Puerto Rico. The second trial was conducted at lake Cerrillos, Ponce.

3.1.1. Results periphytometer study

No significant differences between the different reaction times were observed in the first trial (Figure 3.4). However, on the second trial a significant difference in algae response was observed between the 7-day reaction time and longer reaction times (10 and 15 days) (Figure 3.5). Differences between the two trials may be a result of the robustness of the largest reservoir to exhibit a response to nutrient additions in relation to the small pond. Nevertheless our results indicate that a 10 day reaction time was adequate to characterize aquatic biomass response to nutrients by the Matlock periphytometer approach. On that basis a 10-day reaction time was chosen for subsequent trials.

Figure 3-1. Deployment of the Matlock periphytometer at the Cerrillos lake.

Figure 3-2. Transport of the Matlock periphytometer to the deployment site at the Cerrillos lake.

Figure 3-3. Overview of the algae growth median of different treatments after 7 days of reaction in the Cerrillos lake.

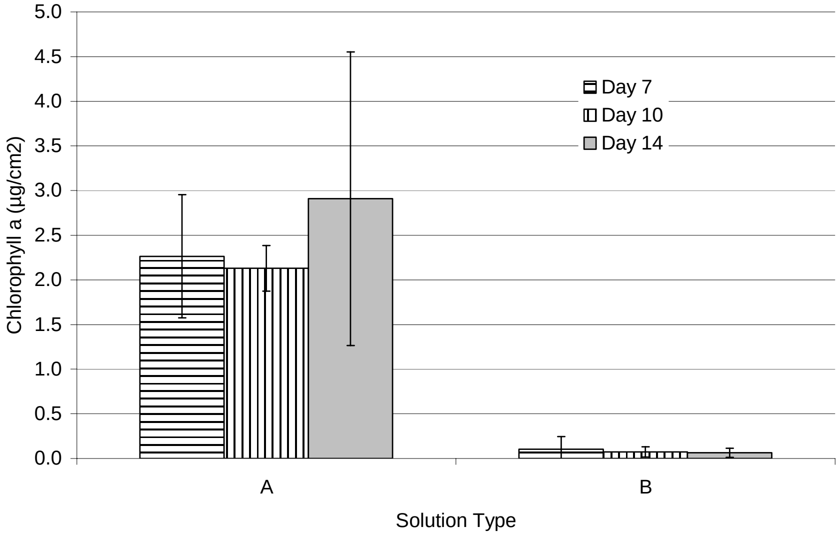

Figure 3-4. Effect of the reaction time on chlorophyll *a* concentrations (periphytometer study). Results from an experimental trial conducted at the Isabela Agricultural Experimental Station (lagoon water chemical status - TKN = 0.39 mg/L, TP = 0.027 mg/L).

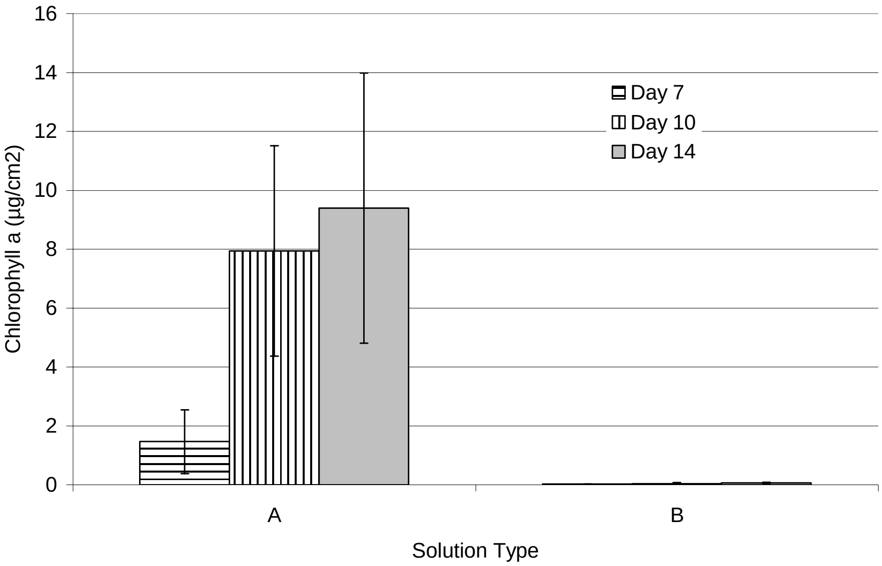

Figure 3-5: Effect of time and solution concentration on Chlorophyll *a* concentrations at Lake Cerrillos. (TKN = 0.41 mg/L, TP = 0.022 mg/L)

3.1.2. Evaluation of limiting nutrient effect on aquatic biomass growth

Once the adequate reaction time was established a series of trials were conducted to evaluate aquatic biomass response to nutrient additions.

The first trial was conducted at Guajataca following the same experimental set up described in the previous experiment. The objective was to identify the nutrient (N and/or P) most limiting to aquatic biomass growth. Results from temperate regions suggest that phosphorus is the controlling factor for aquatic biomass growth on lakes.

Treatments evaluated were:

Treatment Number	**Treatment**
1	-N, -P
2	+N, -P
3	-N, +P
4	+N, +P

where: +N = 14 mg N/L
+P = 2 mg P/L
-N-P = distilled water

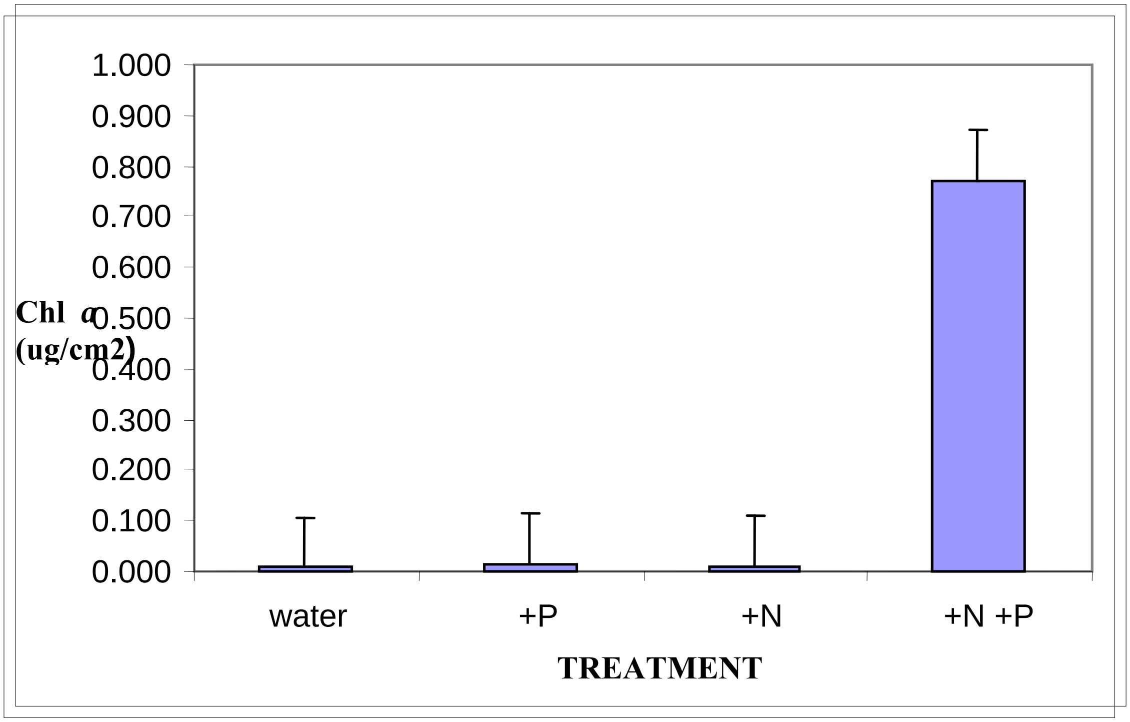

Figure 3-6. Effect of N, P and N+P on primary production in Guajataca Lake.

Table 3-1. Summary of treatment results for trial on nutrient effect on primary production.

Treatment	**Average (Chl. a μ/cm^2)**	**Std. Dev.**
Distilled water	0.008	0.0042
+ P	0.014	0.0142
+ N	0.009	0.0058
+ N, + P	0.771	0.5706

Results indicate that both N, and P are limiting factors to aquatic biomass growth on this lake (Table 3.1, Figure 3.6). Similar results had been previously obtained at Cerrillos (Figure 3.7), although in that case the loss of several experimental units prior to the trial harvest precluded the results to be conclusive. The response pattern observed in our lakes contrasts with results obtained in temperate lakes, where P has been identified as the sole controlling factor to algae growth (Vollenweider, R.A., 1976; Correl, 1998).

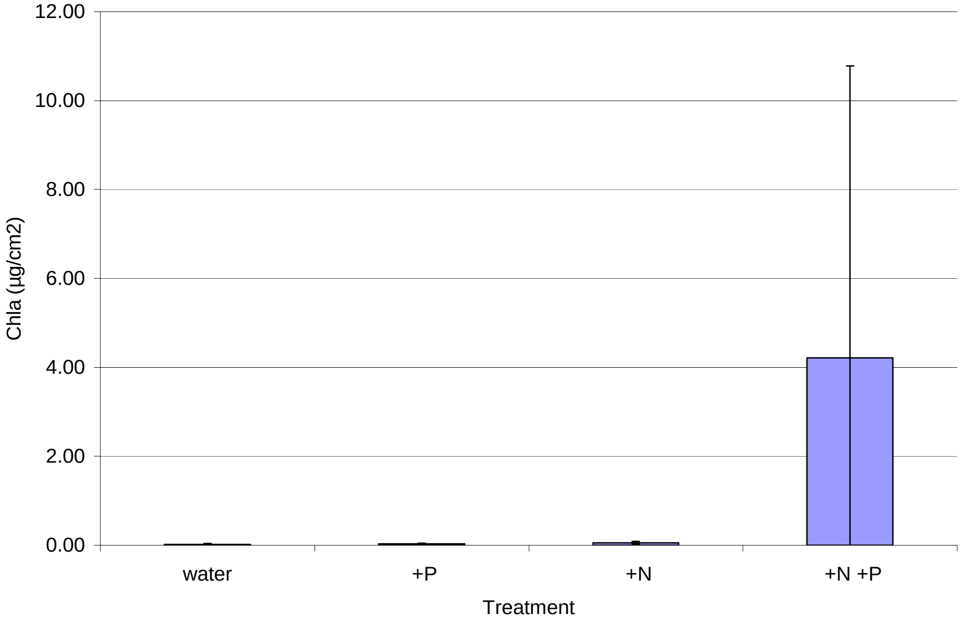

Figure 3-7. Effect of N, P and N+P on primary production in Lake Cerrillos

Following these trails we decided to conduct a series of response curve studies to ascertain if a concentration threshold for each limiting nutrient (N and P) could be established. Initially a trial was conducted to evaluate the effect of variations in P concentrations under non-limiting N conditions. Treatments and schematic description of the experimental set up are presented in Tables 3.2 and 3.4. Results are shown in Tables 3.3, and Figures 3.8 to 3.10.

Table 3-2. Treatment description of P response study.

Treatment No.	**P (mg/L)**	**N (mg/L)**
1	0	14
2	0.02	14
3	0.1	14
4	0.5	14
5	1.0	14
6	2.0	14
7	4.0	14

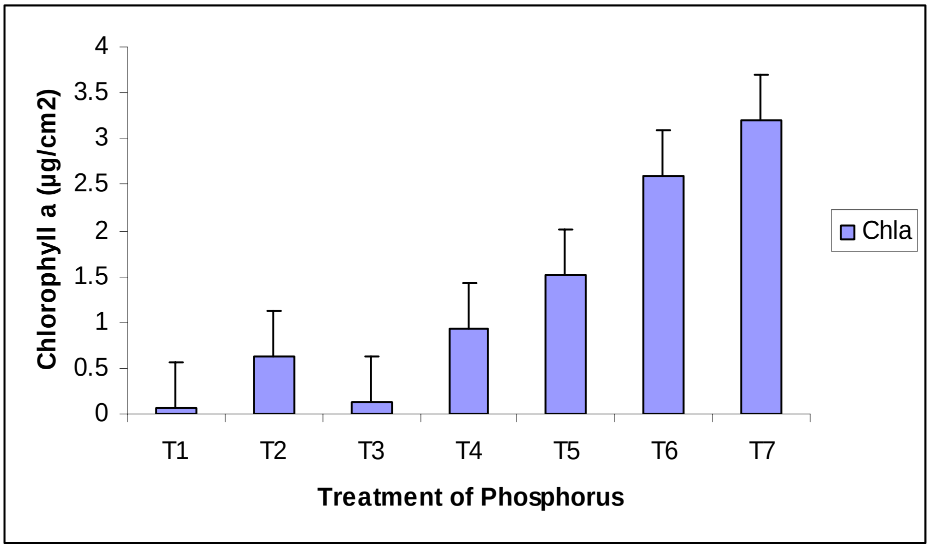

Figure 3-8. Effect of variations in phosphorus concentrations on primary production in Guajataca Lake.

Table 3-3. Descriptive statistics of experimental results (trial A) for P response study.

	Trial A		**Trial B**	
Treatment	**Average Chl *a* µg/cm2**	**Std. Dev.**	**Average Chl *a* µg/cm2**	**Std. Dev.**
T 1	0.062	0.051	0.0338	0.005
T 2	0.625	0.92	0.105	0.05
T 3	0.133	0.066	0.615	0.34
T 4	0.920	1.16	1.130	0.37
T 5	1.504	0.51	1.141	0.93
T 6	2.590	2.05	2.208	0.75
T 7	3.203	1.13	1.803	0.43

The experiment was repeated immediately (October 20, 2004- Trial B) to acquire a sense on the reproducibility of the results. Relatively good reproducibility between trials was obtained (Table 3.3, Figures 3.9, 3.10).

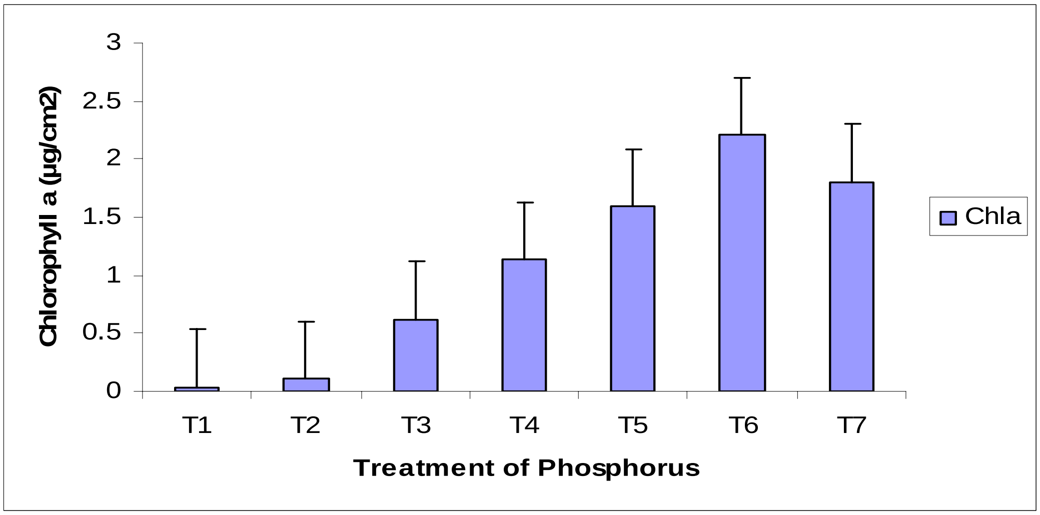

Figure 3-9. Effect of variations in phosphorus concentrations on primary production in Guajataca (trial B).

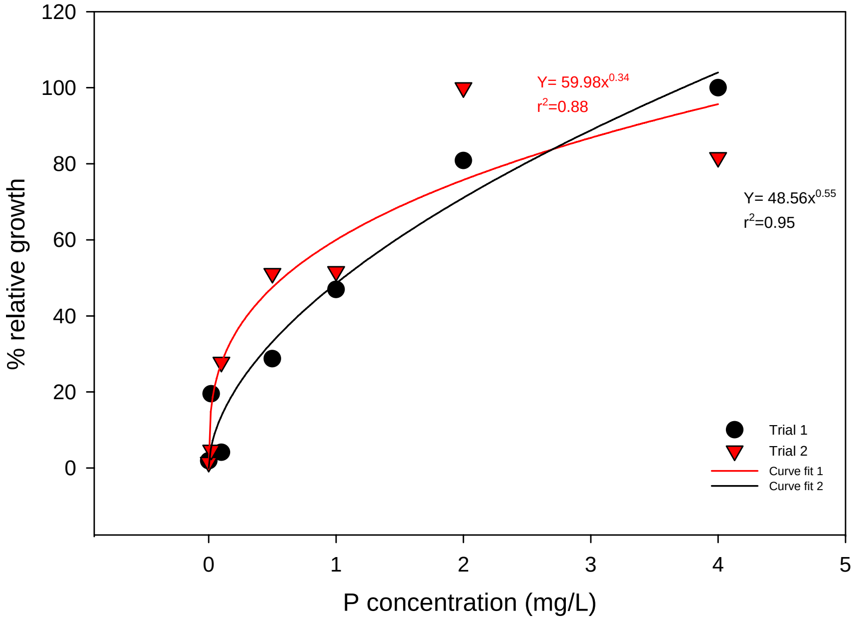

Figure 3-10. Relative response curve of algae growth to P additions. Lines represent empirical descriptions of the experimental points obtained through a curve fitting procedure.

Similar trials were conducted with nitrogen, that is, we evaluated aquatic biomass response to variations in N under non-limiting P conditions. A description of the treatments evaluated is presented in Table 3.4. Results are presented in Table 3.5, and Figures 3.11, and 3.12.

Table 3-4. Description of treatments for the N response trials.

Treatment No.	**P (mg/L)**	**N (mg/L)**
1	2	0
2	2	0.25
3	2	1
4	2	2
5	2	5
6	2	10
7	2	14

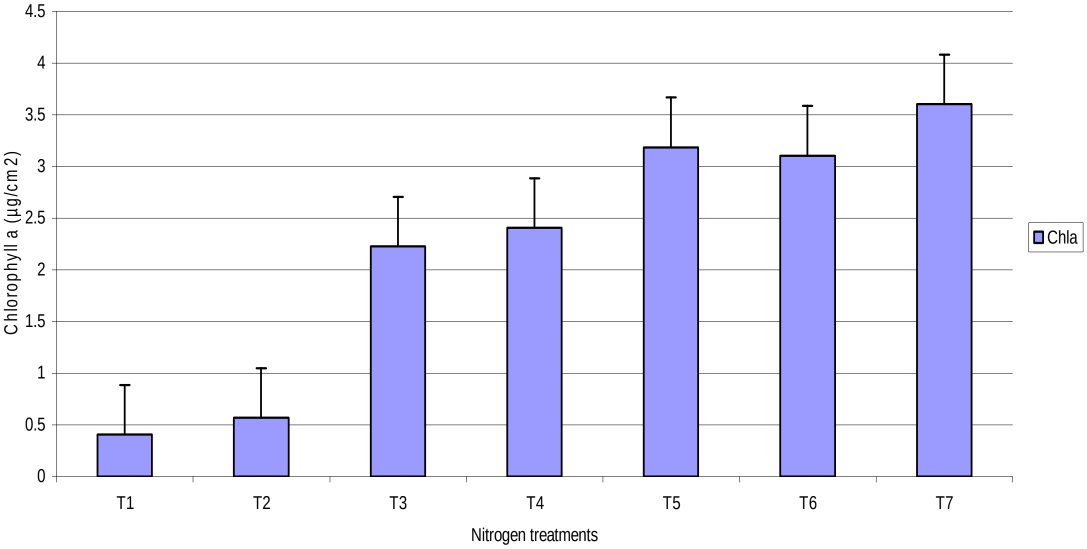

Figure 3-11. Matlock periphytometer study on the effect on nitrogen additions on algae biomass growth at Guajataca. (trial A).

Table 3-5. Descriptive statistics of N response studies.

	Trial A		**Trial B**	
Treatment ID	**Chl *a* mean**	**Std. Dev.**	**Chl *a* mean**	**Std. Dev.**
T1	0.406	0.12	0.37	0.13
T2	0.57	0.17	0.46	0.08

Table 3.5. (cont.)

Treatment ID	**Chl *a* mean**	**Std. Dev.**	**Chl *a* mean**	**Std. Dev.**
T3	2.23	0.45	0.52	0.05
T4	2.41	0.78	1.11	0.85
T5	3.19	0.38	1.49	1.13
T6	3.11	1.08	2.04	1.13
T7	3.60	0.90	1.89	1.05

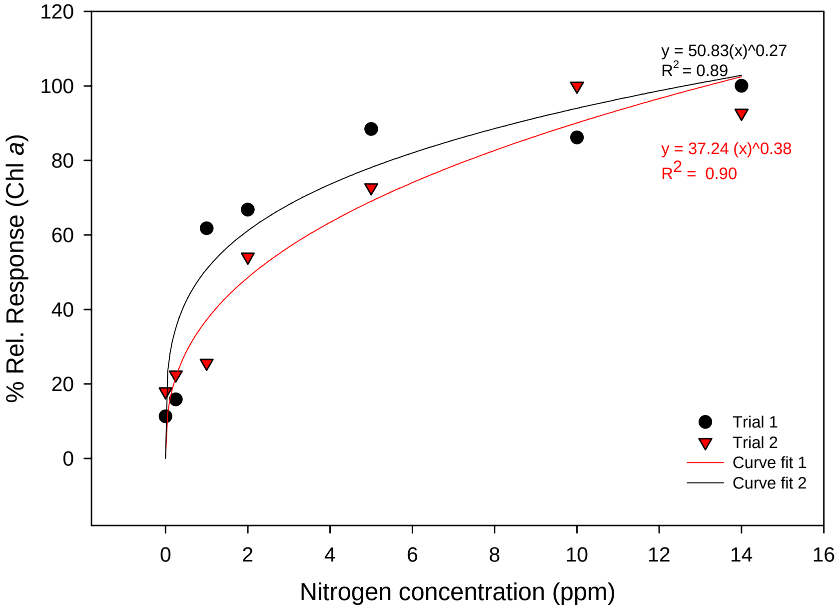

Figure 3-12. Relative response curve of algae growth to N additions. Lines represent empirical descriptions of the experimental points obtained through a curve fitting procedure.

Results demonstrate the importance of controlling nutrient loadings (both N and P) into reservoirs of Puerto Rico. Our experimental approach proved to be an adequate qualitative tool for making inferences on nutritional conditions limiting algae growth in reservoirs. However, after much consideration we felt the approach was not sensitive enough to identify threshold concentrations representing reference conditions. Thus, a systematic monitoring program was implemented at the Guajataca reservoir to evaluate phytoplankton response to seasonal changes in the reservoirs nutritional content. Our hypothesis was that, this being an oligotrophic-mesotrophic reservoir, phytoplankton community structural changes occurring as result of variations in nutritional status could potentially be used to establish a biological index of reference conditions applicable to other reservoirs of the island.

3.2 GUAJATACA MONITORING STUDY

The monitoring program consisted on a weekly scheduled sampling at the two PREQB Guajataca lake sampling stations [i.e., 10720 (lake entrance); 10790 (lacustrine zone)] (Figure 2.13). Sampling began on March 2, 2005 and finished at the end of July. Samples were collected at 0m (surface), 1m, and at 5m (below the photic zone (determined by secchi depth)] with a Van Dorn sampler and analyzed for TKN, TP, Chl *a*, and DOC (Appendix C). In addition, the following parameters were determined *in-situ:* pH, electrical conductivity, dissolved oxygen, temperature (YSI 85 multiparameter meter), secchi depth and turbidity. A characterization of phytoplankton diversity on the different samples was performed according to different specialized keys for tropical and subtropical areas (Appendix E) (Prescott, et al., 1970; Parra, et al., 1982).

Figure 2-13. Location of sampling stations used in our monitoring program: 1) Entrance (USGS 50010720), 2) Dam (USGS 50010790).

3.2.1. Results – nutrient monitoring study

Figures 3.14 through 3.16 show the precipitation, water level registered at the reservoir and a relation of the sampling events during the sampling period. After a relatively dry period from February until the end of March, significant amounts of precipitation occurred during the months of April and May. June was a relatively dry month, whereas a July began with an unusual storm event in which 26.17 inches of rain were recorded within a 24 hour period. After this date (July 1st) the rainfall pattern normalized remaining relatively wet for the remainder of the sampling season.

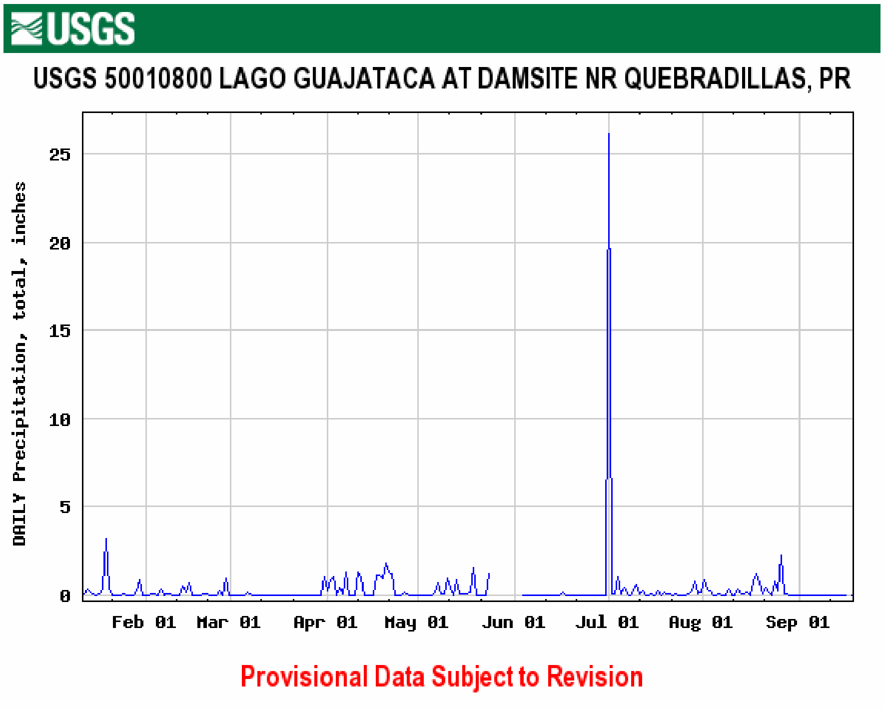

Figure 3-14. Precipitation recorded at Guajacata (USGS 50010800 station (dam)) during the sampling season.

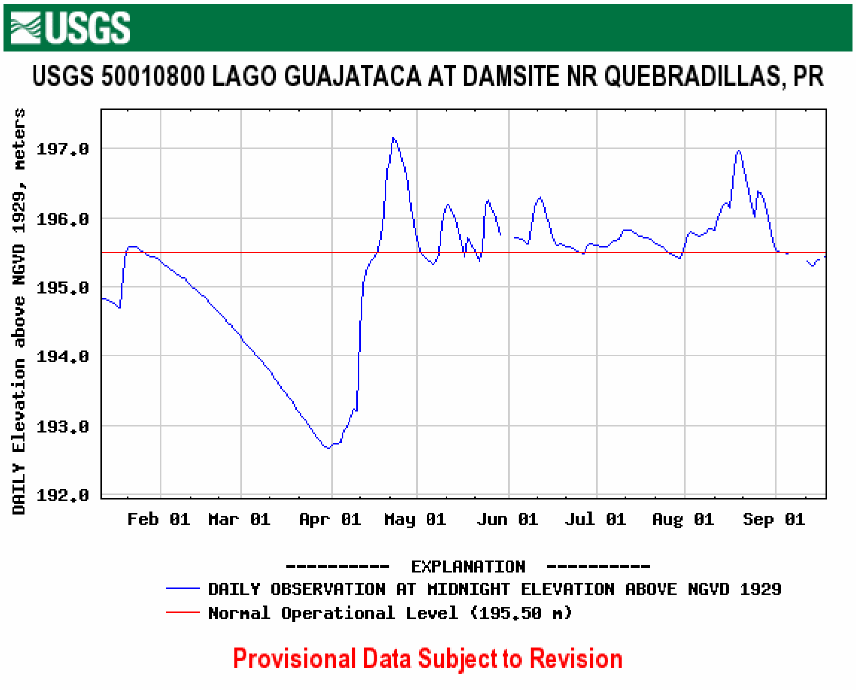

Figure 3-15. Reservoir's elevation during the study period (dam station -USGS 50010800).

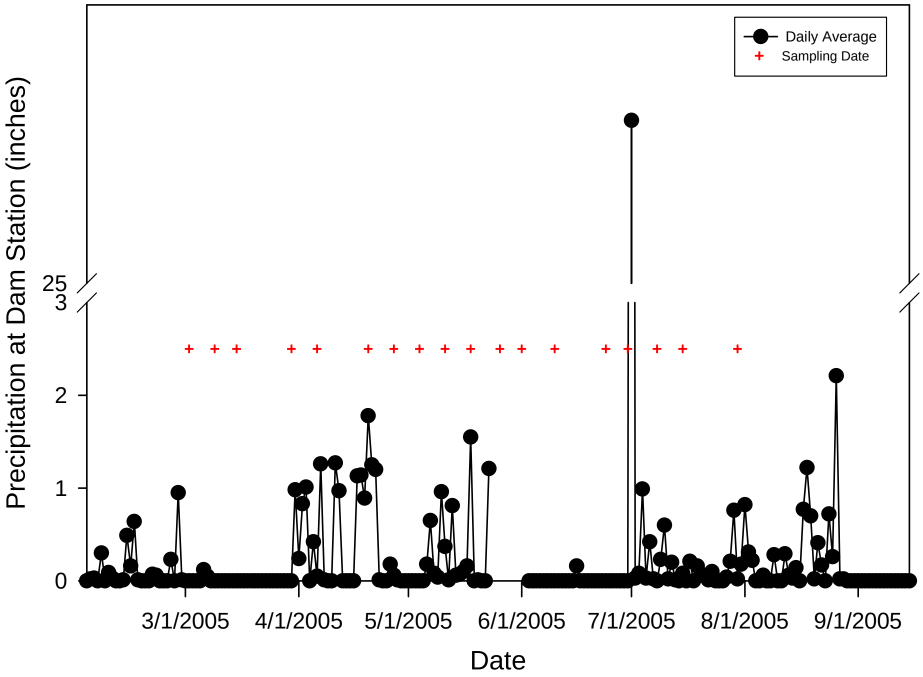

Figure 3-16. Relation between rainfall pattern and sampling events at Guajataca. Sixteen sample events were conducted between March 1 and July 30th 2005. Results for the chemical characterization of the collected samples are shown in Tables 3.6, and 3.7 (the complete data set is included in Appendix 1).

Table 3-6. Characterization of samples from the Guajataca study.

Date[1]	Station No.	Depth[2]	Chl a ug/l	TKN mg/L	TP mg/ L	DOC mg/L	TEM P ˚C	DO mg/L	COND µS	pH	S.S mg/ L
3/2/05	1	1	1.02	0.179	0.013	5.37	27.30	8.15	265.10	7.81	3.2
3/2/05	1	2	2.11	0.206	0.018	4.23	26.40	6.58	273.40	7.80	7.6
3/2/05	1	3	3.80	0.215	0.015	4.92	24.10	6.10	281.60	7.70	9.2
3/2/05	2	1	0.94	0.132	0.007	3.85	27.50	7.82	291.70	7.90	1.6
3/2/05	2	2	1.09	0.160	0.011	3.92	26.00	7.50	276.20	7.70	2.4
3/2/05	2	3	1.42	0.139	0.009	4.37	24.70	6.79	243.90	7.68	3.2
3/9/05	1	1	1.28	0.225	0.019	4.02	27.30	6.07	302.00	7.89	2.8
3/9/05	1	2	3.09	0.201	0.018	3.64	26.90	5.85	308.90	7.71	6.8
3/9/05	1	3	0.84	0.367	0.075	4.60	26.60	4.26	351.60	7.33	13.6
3/9/05	2	1	0.51	0.147	0.012	3.26	27.30	7.62	303.70	7.98	2.4
3/9/05	2	2	1.02	0.147	0.013	3.55	26.40	8.20	299.40	7.96	22.8
3/9/05	2	3	1.12	0.210	0.011	3.43	25.80	8.80	296.40	7.93	1.2
3/15/05	1	1	1.43	0.207	0.018	3.23	27.60	8.30	295.10	8.12	1.6
3/15/05	1	2	1.54	0.223	0.020	3.35	27.50	7.70	296.50	7.70	3.4
3/15/05	1	3	1.36	0.211	0.007	3.41	27.60	4.37	296.80	8.02	5.6
3/15/05	2	1	0.48	0.224	0.017	3.32	27.50	8.45	281.50	8.16	1.8
3/15/05	2	2	0.84	0.128	0.017	3.34	28.40	5.40	133.10	8.22	3.8
3/15/05	2	3	2.97	0.155	0.013	3.54	26.00	3.25	157.10	7.62	4.0
3/30/05	1	1	3.00	0.567	0.030	1.33	28.90	7.40	301.60	8.17	1.0
3/30/05	1	2	3.90	0.522	0.038	1.41	27.60	7.30	298.40	7.85	1.6
3/30/05	1	3	6.24	0.427	0.036	1.16	26.80	3.90	318.50	7.81	3.0
3/30/05	2	1	3.85	0.338	0.025	1.23	29.00	7.60	300.10	8.06	1.2
3/30/05	2	2	9.20	0.368	0.032	1.32	27.90	7.83	287.90	8.02	1.0
3/30/05	2	3	3.07	0.341	0.025	1.29	28.70	4.82	280.70	8.03	3.2
4/6/05	1	1	9.40	0.413	0.036	0.91	27.50	8.60	267.60	8.35	1.2
4/6/05	1	2	9.49	0.469	0.040	1.16	27.50	8.40	283.50	8.10	2.0
4/6/05	1	3	4.42	0.547	0.038	1.61	27.30	4.35	248.40	7.69	6.0
4/6/05	2	1	0.47	0.333	0.043	0.92	28.20	8.18	289.40	8.30	1.2
4/6/05	2	2	5.33	0.346	0.022	2.54	27.90	7.81	289.10	8.24	1.2
4/6/05	2	3	5.68	0.317	0.024	2.39	27.40	3.45	171.60	7.69	2.0
4/20/05	1	1	7.51	0.432	0.042	3.46	28.50	10.30	248.50	8.46	1.4
4/20/05	1	2	8.75	0.474	0.054	3.23	28.40	9.85	232.70	8.49	2.2
4/20/05	1	3	3.71	0.431	0.050	5.13	26.80	6.48	264.10	7.83	10.2
4/20/05	2	1	5.15	0.429	0.027	3.29	27.50	8.45	262.70	8.29	1.8
4/20/05	2	2	5.20	0.363	0.023	3.19	27.10	8.41	248.40	8.27	1.2
4/20/05	2	3	5.45	0.358	0.025	3.14	26.90	1.32	236.50	8.25	1.0
4/27/05	1	1	29.48	0.615	0.072	4.80	28.50	11.82	260.60	8.63	1.1
4/27/05	1	2	25.27	0.642	0.071	5.03	27.80	7.22	264.50	8.72	1.6
4/27/05	1	3	7.82	0.445	0.052	4.77	25.80	1.84	128.70	7.71	3.2
4/27/05	2	1	5.22	0.450	0.026	3.53	29.20	9.11	284.50	8.62	1.4
4/27/05	2	2	5.27	0.427	0.026	3.55	28.60	8.36	283.10	8.64	1.0
4/27/05	2	3	7.58	0.436	0.025	4.09	26.90	2.98	149.20	7.65	1.2
5/4/05	1	1	6.85	0.440	0.043	4.04	29.00	8.21	247.90	8.54	3.6
5/4/05	1	2	7.63	0.445	0.038	4.15	28.60	7.73	265.80	8.39	3.2

Table 3.6 (cont.)

Date[1]	Station No.	Depth[2]	Chl a ug/l	TKN mg/L	TP mg/L	DOC mg/L	TEMP ˚C	DO mg/L	COND µS	pH	S.S mg/L
5/4/05	1	3	2.25	0.291	0.026	3.79	26.00	1.90	331.70	7.26	4.0
5/4/05	2	1	4.63	0.312	0.017	1.48	29.20	8.06	271.90	8.49	3.6
5/4/05	2	2	3.99	0.404	0.015	4.67	29.00	7.68	272.20	8.39	3.2
5/4/05	2	3	5.36	0.429	0.024	4.56	26.80	2.80	310.50	7.70	4.8
5/11/05	1	1	11.52	0.553	0.033	3.21	29.50	8.67	260.50	8.64	3.6
5/11/05	1	2	102.91	1.218	0.125	4.03	29.00	9.58	254.40	8.65	8.0
5/11/05	1	3	20.90	0.684	0.068	3.86	26.50	3.59	286.50	7.70	9.6
5/11/05	2	1	13.41	0.436	0.017	3.10	29.50	7.50	260.50	8.41	1.2
5/11/05	2	2	7.34	0.408	0.020	3.77	28.80	7.37	257.60	8.36	0
5/11/05	2	3	89.57	0.492	0.017	3.11	26.80	0.53	311.70	7.51	1.2
5/18/05	1	1	43.52	0.854	0.066	3.85	27.20	6.01	253.00	7.79	8
5/18/05	1	2	30.99	0.630	0.051	4.04	27.20	5.96	252.70	7.65	2.4
5/18/05	1	3	6.59	0.610	0.025	3.49	26.90	0.94	288.30	7.35	3.2
5/18/05	2	1	46.37	0.404	0.039	4.01	27.60	6.89	254.00	8.30	1.2
5/18/05	2	2	23.59	0.536	0.023	3.45	27.50	7.05	254.00	8.03	3.6
5/18/05	2	3	11.71	0.444	0.023	3.69	26.80	0.84	303.10	7.69	2.4
5/26/05	1	1	5.78	0.590	0.045	3.60	30.60	9.58	238.10	8.50	4.4
5/26/05	1	2	34.38	0.934	0.091	3.77	29.10	7.30	232.70	8.35	8.4
5/26/05	1	3	3.61	0.480	0.045	4.62	26.80	1.71	275.90	7.45	7.2
5/26/05	2	1	25.32	0.738	0.033	3.58	31.20	6.90	225.30	8.48	10.4
5/26/05	2	2	36.36	0.704	0.037	3.84	29.00	7.46	239.90	8.40	8.8
5/26/05	2	3	3.77	0.412	0.022	3.75	26.50	0.18	297.80	7.37	1.6
6/1/05	1	1	8.30	0.499	0.032	3.66	30.30	7.38	231.30	8.56	2.4
6/1/05	1	2	32.22	0.825	0.078	4.03	29.70	7.76	228.90	8.45	0.8
6/1/05	1	3	7.31	0.496	0.037	4.31	26.70	0.18	300.50	7.32	1.2
6/1/05	2	1	2.82	0.507	0.017	3.51	30.60	6.56	233.50	8.43	0
6/1/05	2	2	4.12	0.416	0.022	3.33	30.00	6.96	226.10	8.31	1.6
6/1/05	2	3	4.61	0.330	0.025	5.02	26.30	2.10	311.60	7.25	0
6/10/05	1	1	8.70	0.369	0.040	2.69	29.80	7.15	260.00	8.54	1.2
6/10/05	1	2	46.56	0.447	0.068	3.12	29.00	6.76	255.40	8.01	6.8
6/10/05	1	3	5.70	0.304	0.045	2.98	27.20	2.49	277.00	7.77	10
6/10/05	2	1	3.49	0.283	0.034	2.73	30.10	6.89	236.90	8.44	1.6
6/10/05	2	2	6.01	0.340	0.044	3.41	29.10	5.23	235.90	8.28	1.2
6/10/05	2	3	8.52	0.371	0.035	3.13	27.00	1.30	306.90	7.36	4.4
6/24/05	1	1	19.32	0.430	0.057	2.77	29.80	7.64	241.50	8.31	1.6
6/24/05	1	2	21.84	0.528	0.073	3.11	29.70	6.44	241.00	8.25	0.8
6/24/05	1	3	6.15	0.342	0.049	3.10	28.20	2.90	292.80	7.78	2.8
6/24/05	2	1	2.71	0.303	0.038	2.69	30.10	6.67	221.40	8.23	4.0
6/24/05	2	2	4.00	0.283	0.029	2.50	30.20	6.17	243.80	8.09	4.0
6/24/05	2	3	4.89	0.321	0.039	2.77	29.30	2.10	247.60	7.80	3.2
6/30/05	1	1	4.57	0.347	0.043	2.89	29.40	6.90	224.80	8.63	3.6
6/30/05	1	2	6.47	0.379	0.043	3.07	29.30	5.85	225.50	7.72	6.4
6/30/05	1	3	3.83	0.388	0.053	3.25	28.20	2.45	273.20	7.50	6.0
6/30/05	2	1	3.16	0.315	0.032	2.75	30.00	7.88	248.00	8.50	0.8

Table 3.6 (cont.)

Date[1]	**Station No.**	**Depth**[2]	**Chl a ug/l**	**TKN mg/L**	**TP mg/L**	**DOC mg/L**	**TEMP ˚C**	**DO mg/L**	**COND µS**	**pH**	**S.S mg/L**
6/30/05	2	2	5.81	0.354	0.038	2.91	29.70	6.78	245.30	8.36	6.4
6/30/05	2	3	6.71	0.355	0.036	2.95	29.10	3.94	255.20	7.67	4.0
7/8/05	1	1	8.69	0.380	0.044	3.00	28.90	7.61	255.80	8.41	13.6
7/8/05	1	2	8.62	0.420	0.043	2.90	28.90	7.23	256.50	8.48	1.6
7/8/05	1	3	4.46	0.254	0.036	2.92	28.80	3.70	257.10	7.96	1.2
7/8/05	2	1	5.35	0.315	0.033	2.78	30.00	7.41	253.20	8.59	1.6
7/8/05	2	2	5.65	0.353	0.036	2.81	29.80	6.89	252.80	8.66	0.4
7/8/05	2	3	5.89	0.338	0.039	3.06	28.50	2.40	366.10	7.56	28.4

1 – Dates are expressed as: day/month/year

2 – Depth refers to: 1 = surface (0M); 2 = 1m; 3 = photic zone

Table 3-7: Statistical summary of samples obtained in the Guajataca study. Refer to Appendix D for complete statistical report.

Parameter	Chl a µg/L	TKN mg/L	TP mg/L	DOC mg/L	Temp. ºC	DO mg/L	Cond. µS/cm	pH	S.S. mg/L
Average	10.65	0.406	0.035	3.31	28.09	6.07	263.60	8.06	3.94
Mediann	5.41	0.384	0.033	3.35	28.05	6.90	261.65	8.08	2.60
St. Dev.	16.23	0.179	0.020	0.98	1.40	2.59	39.72	0.40	4.36
Min.	0.47	0.128	0.007	0.91	24.10	0.18	128.70	7.25	0.00
Max.	102.91	1.218	0.125	5.37	31.20	11.82	366.10	8.72	28.40

Based on the average TP and Chl *a* concentrations this reservoir could be classified as mesotrophic-eutrophic, which contrasts with our original assumption that this was an oligothrophic-mesotrophic lake. Apparently, the rainfall events that predominated the study period incremented the nutrients load to this lake to a point that may have affected its nutritional status. A statistical evaluation of the results revealed significant differences between stations for TP, TKN, DOC, DP, turbidity, and transparency (Table 3.8), (Appendix D). Higher nutrient concentrations (TKN, TP, DP, DOC) were observed in station 1 (riverine) in comparison to station 2 (lacustrine). Although not significantly different, both chlorophyll a and suspended sediments were higher in station 1. Turbidity was also higher in station 1. As a result, transparency was higher in station 2. This coincides with the general behavior observed in reservoirs where higher nutrient and sediment loadings are generally observed at the entrance.

Table 2-8. Statistical comparison between stations (riverine and lacustrine) at Guajataca (all sampling depths combined).

Para-meter	Chl a (µg/L)	DO (mg/L)	SS (mg/L)	TKN (mg/L)	TP (mg/L)	DP (mg/L)	DOC (mg/L)	Turb-idity	Trans-parency
Station 1	12.32a	6.20a	4.37a	0.46a	0.045a	0.015a	3.44a	4.63a	1.73b
Station 2	8.08a	5.94a	3.26a	0.35b	0.025b	0.0096b	3.18b	2.27b	2.17a
MSD*	4.88	0.50	1.49	0.04	0.006	0.0031	0.197	1.01	0.21

Both TKN, and TP exhibited significant differences between sampling dates, with the lowest concentrations occurring in March and the highest occurring between mid-May and June. This coincides with the precipitation and water level pattern recorded at the dam station of the lake (Figures 3.14 and 3.15).

In general no significant differences were observed between sampling depths for either nutrient (Table 3.9). Dissolved oxygen did showed a significant drop in concentration below the 5m depth. Average concentration fall well below the 5mg/L criteria established by USEPA. These results are similar to those reported by Townsend (1999) for two tropical reservoirs of Australia. The author indicated that tropical waters are more susceptible to oxygen depletion than their temperate counterparts. He attributed this behavior to the reduced solubility of oxygen in warm waters coupled with higher rates of microbial metabolism. The effect is compounded by the long periods of stratification commonly exhibited by some tropical reservoirs, which causes isolation of hypolimnetic waters from surface waters oxygenated by photosynthesis and surface-air gas exchange. In recent years researchers have emphasized that hypolimnetic anoxia is a temperature dependent process which is not exclusively related to organic loads. As a result they recommend discontinue using hypolimnion anoxia as an indicator of the trophic status of tropical lakes/reservoirs as commonly used in the temperate regions.

Table 3-9: Difference between sampling depths (overall) at Guajataca

Para-meter	Chl a (µg/L)	DO (mg/L)	SS (mg/L)	TKN (mg/L)	TP (mg/L)	DOC (mg/L)	Turb-idity	E.C. dS/cm	pH
0m	8.78a	7.79a	2.81a	0.40a	0.033a	3.09a	2.48b	260.14a	8.32a
1m	14.28a	7.20a	3.81a	0.45a	0.040a	3.32a	3.28ab	255.81a	8.17b
5m	7.53a	3.21b	4.83a	0.37a	0.033a	3.51a	4.49a	270.44a	7.65c
MSD	8.63	0.80	2.3	0.11	0.012	0.58	1.87	21.12	0.15

A correlation analysis revealed some important trends (Tables 3.10 and 3.11). A strong positive correlation was observed between concentrations of TKN and TP. This was especially evident at the entrance, which suggests that similar transport factors are controlling the loadings of these nutrients into the reservoir. The relationship between TKN and TP weakens at the dam section of the lake. This is probably due to differences in the mechanisms (e.g., atmospheric deposition, biotic and abiotic losses, transformation reactions, etc), that control each nutrient's (i.e., TKN, and TP) budget within a reservoir system. There was a strong correlation between Chl *a* and nutrients (TKN, and TP) (Figure 2.16 and 2.17). Once again this illustrates the role of nutrients on aquatic biomass response. The relationship is stronger at the lake entrance where nutrient levels were higher. In addition, the relationship drops significantly at the 5m depth evidencing the importance of the photoactive zone in regulating phytoplankton activity in reservoir systems.

An interesting relationship was observed between the ratio of TN/TP (i.e., (TKN + NO_3)/TP) and TP (Figure 3.18). The relationship results in a first order decay function whose change point on the TP axis could be regarded as a transition point between the phosphorus-limiting, and the nitrogen-limiting zones. This relationship can be represented by two straight lines (equations 3, and 4) that merge together at a change point that defines two populations.

Below the change point: $y = m_1x + c$ [3]

Above the change point: $y = m_1x + m_2(CP) + c$ [4]

Where: c is the intercept; m_1 is the slope of the linear relationship for the range of x values less than the change point; m_2 is the difference in slopes after the change point compared to m_1; and CP is the change point.

The slopes of these lines are significantly different from each other. A discontinuous regression model was applied to estimate the breakpoint (the point that defines a change in the relationship between variables). Specifically, the Nonlinear breakpoint regression model in the STATISTICA® software was used. The model employs a *quasi-Newton* algorithm that approximates the second order derivative of the loss function to estimate the best fitting parameters. For this data set the estimated change point was 17.10 µg/L (Figure 3.19), a value remarkably similar to the reference value for TP proposed in this study (17.0 µg/L).

Table 3-10: Pearson correlation coefficients for selected parameters

Parameters	Overall	Station 1	Station 2
Chl a – TKN	0.69	0.80	0.50
Chl a – TP	0.57	0.80	0.11
TKN – TP	0.74	0.81	0.40
TKN – DP	0.63	0.73	0.27
TP- DP	0.80	0.82	0.76
Temp – pH	0.62	0.62	0.62
DO – pH	0.74	0.79	0.70
Turbidity-Transparency	-0.50	-0.47	-0.62
Turbidity-Suspended solids	0.31	0.48	0.06

Table 2-11. Pearson correlation coefficients for samples obtained at different depths

Parameters	**Riverine station**			**Lacustrine station**		
	0m	**1m**	**5m**	**0m**	**1m**	**5m**
Chl a – TKN	0.77	0.84	0.67	0.50	0.85	0.48
Chl a – TP	0.83	0.89	0.38	0.36	0.38	-0.15
TKN – TP	0.74	0.94	0.44	0.34	0.45	0.41
TKN – DP	0.62	0.90	-0.02	0.14	0.43	0.27
TP- DP	0.89	0.95	0.25	0.59	0.86	0.92
Temp – pH	0.65	0.41	0.04	0.54	0.44	0.26
DO – pH	0.45	0.62	0.50	-0.005	0.13	0.40
Turbidity-Transparency	-0.47	--	--	-0.62	--	--
Turbidity-Suspended solids	-0.14	0.35	0.77	0.38	0.30	-0.16

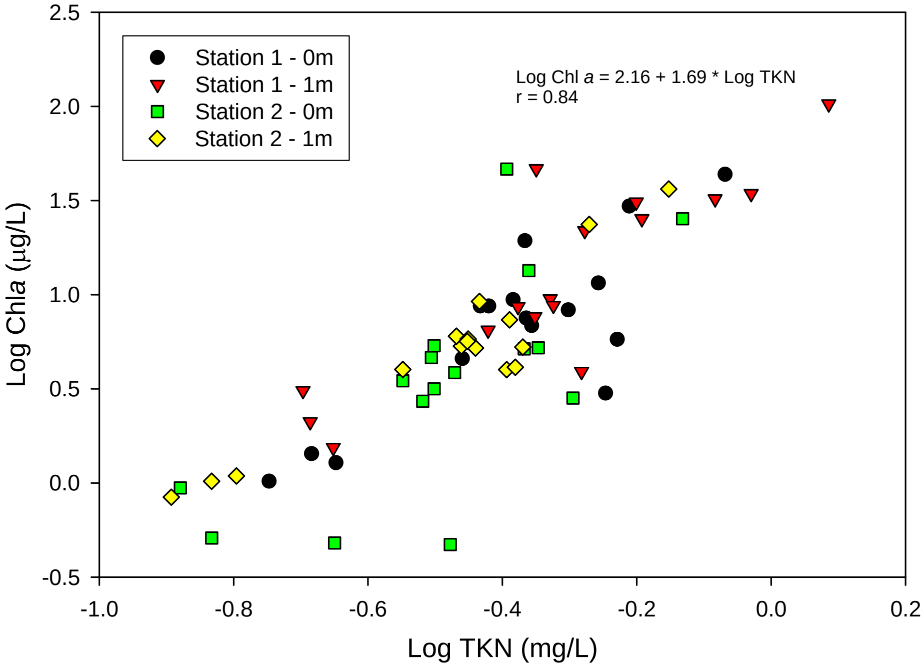

Figure 3-17: Relation between TKN and chlorophyll values for surface samples (0m and 1m) collected at Guajataca during the monitoring study.

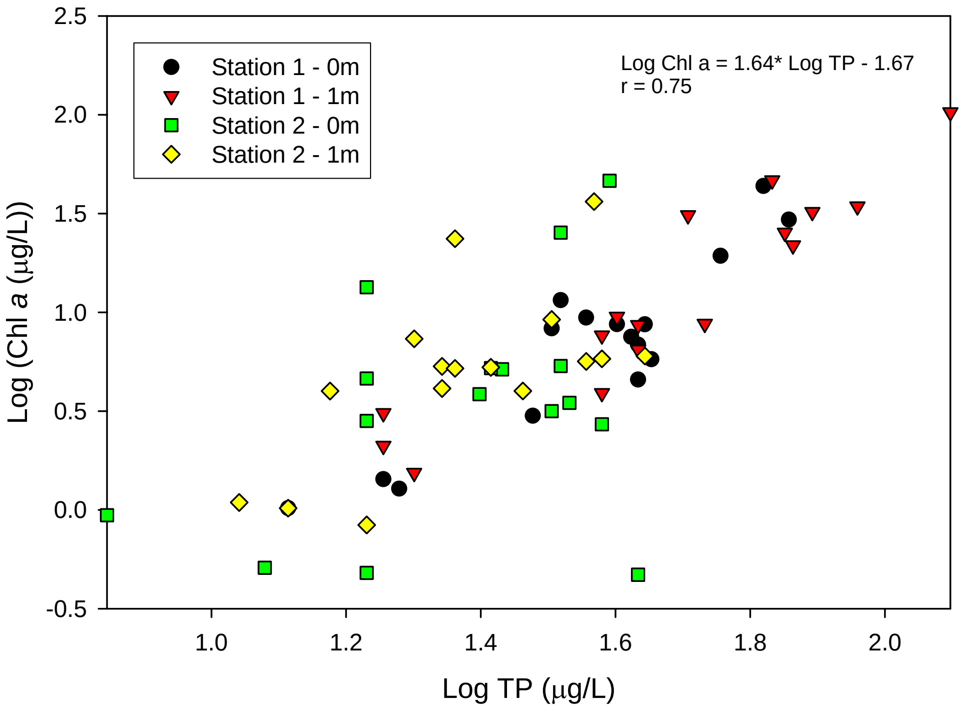

Figure 3-17: Relation between TP and chlorophyll values for surface samples (0m and 1m) collected at Guajataca during the monitoring study.

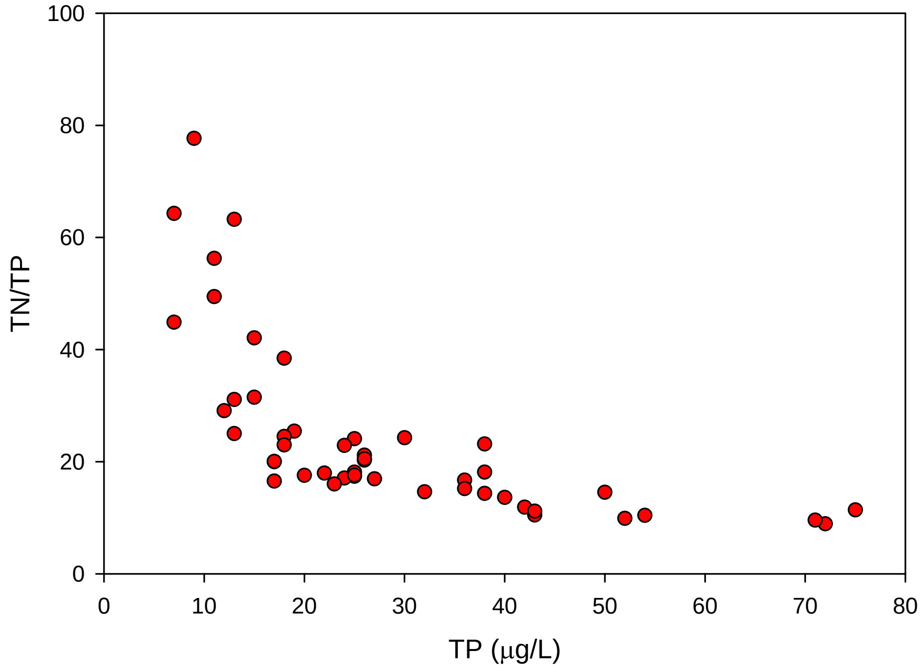

Figure 3-18: Relation between the ratio of TN/TP and TP for samples collected at Guajataca during the monitoring study.

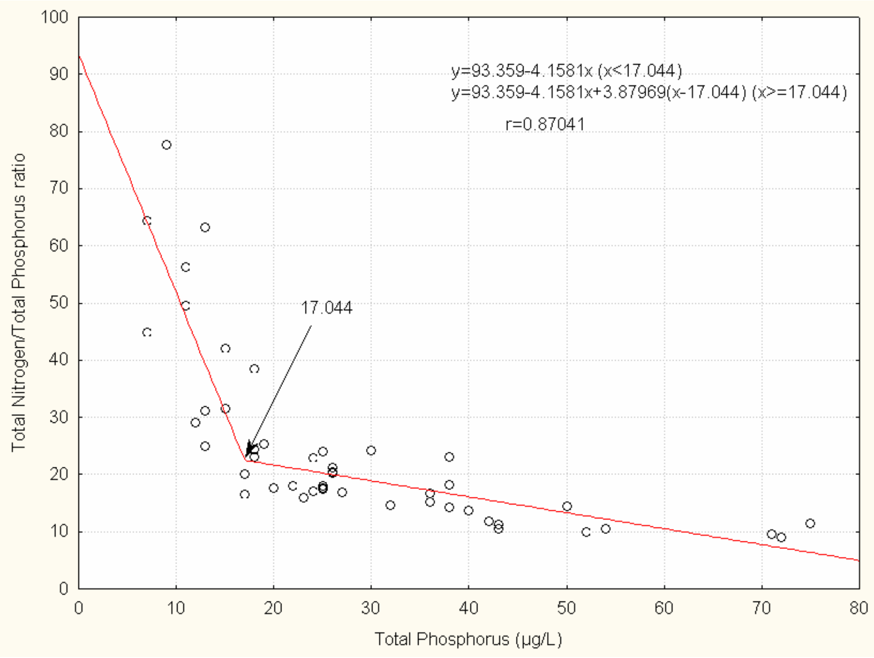

Figure 3.19: Split-line model representation of the relationship between (TP/TN) and TP. The intersection point between the two lines (TP = 17.04) is the change point for the relation.

3.2.2. Phytoplankton diversity

A characterization of the phytoplankton diversity in the lake during the study period revealed two dominant divisions: Cyanophytae, and Pyrrophytae (Table 3.12, Appendix E). A large number of the cyanobacter species (*Microcystis, Aphanothece*, etc.) have been detected in eutrophic waters. It seems that these organisms are more prevalent under these conditions than in oligotrophic conditions (Margalef, 1983). This may be due to their capacity to develop in low CO_2 waters, typical of systems with high algae density as a result of nutrient enrichment. In addition their photosynthetic pigments make them highly efficient in terms of light use reducing the penetration of light to deeper profiles where chlorophytaes may develop (Peinador, 1999). The predominance of Cyanophytas could be indicative of a decline in the quality of the water at this lake. Some cyanobacter genera are potentially hazardous to humans and thus, it is important to prevent conditions that promote their reproduction. Chlorophytaes, Chrysophytaes and Euglenophytates appeared in lesser numbers. Chlorophytaes, and Chrysophytaes are characteristic of mesotrophic systems. Chrysophytaes are typically found in neutral to slightly alkaline soft waters with low to moderate productivity. On the other hand Euglenophytaes have generally been associated with eutrophic to hypereutrophic conditions with a high content of sediments and organic matter.

The composition of the phytoplankton community was relatively similar at both stations (entrance and dam). Only for the chlorophytae division, a larger number of individuals were observed at the entrance (station 1) than at the dam station (Table 3.13). In general the number of algae species were greater at the top portions of the water column (0m, and 1m), than at the lower sampling depth (5m) (Appendix F). The exception was the Chrysophytae division, which showed a greater number of individuals at the greater depth (Table 3.14).

The total number of species was lower at the initial stage of the study, coinciding with the relatively dry period. This effect was more drastic for the Chlorophytae and the Pyrrophytae divisions. As the rainy period started an increase in the number of species was observed (Figure 3.20).

Table 3-12. Characterization of phytoplankton community structure at Guajataca.

			Number of Individuals per Algae Division				
Station No	**Depth[1]**	**Sampling Date**	**Chloro-phytae**	**Cyano-phytae**	**Pyrro-phytae**	**Eugleno-phytae**	**Chryso-phytae**
1	1	3/2/05	15	171	108	12	2
1	2	3/2/05	14	132	48	23	9
1	3	3/2/05	2	14	40	3	13
2	1	3/2/05	10	79	55	7	0
2	2	3/2/05	16	103	27	0	0
2	3	3/2/05	0	6	14	6	4
1	1	3/9/05	12	134	121	9	3
1	2	3/9/05	18	154	85	7	3
1	3	3/9/05	7	6	32	1	6
2	1	3/9/05	3	54	68	4	0
2	2	3/9/05	20	98	43	6	0
2	3	3/9/05	1	16	29	7	5
1	1	3/15/05	7	87	83	6	0
1	2	3/15/05	21	179	79	8	2
1	3	3/15/05	3	11	41	8	11
2	1	3/15/05	6	75	43	2	0
2	2	3/15/05	9	126	30	2	0
2	3	3/15/05	7	11	29	3	0
1	1	3/30/05	6	77	77	6	0
1	2	3/30/05	12	181	68	11	7
1	3	3/30/05	9	5	24	2	6
2	1	3/30/05	4	87	51	0	0
2	2	3/30/05	11	115	29	1	0
2	3	3/30/05	4	5	23	4	3
1	1	4/6/05	23	85	165	3	6
1	2	4/6/05	29	70	123	35	3
1	3	4/6/05	7	41	31	27	18
2	1	4/6/05	17	80	34	0	11
2	2	4/6/05	25	87	85	11	22
2	3	4/6/05	3	10	21	16	44
1	1	4/20/05	39	70	68	13	0
1	2	4/20/05	65	102	79	19	0
1	3	4/20/05	9	29	17	6	7
2	1	4/20/05	23	66	139	9	6
2	2	4/20/05	31	83	143	5	16
2	3	4/20/05	7	32	29	13	18
1	1	4/27/05	67	130	130	5	13
1	2	4/27/05	38	89	106	23	9

Table 3.12. (cont.)

Station No	Depth[1]	Sampling Date	Chloro-phytae	Cyano-phytae	Pyrro-phytae	Eugleno-phytae	Chryso-phytae
1	3	4/27/05	20	35	25	13	3
2	1	4/27/05	8	79	123	4	1
2	2	4/27/05	37	79	110	4	4
2	3	4/27/05	11	60	50	5	30
1	1	5/4/05	51	71	139	4	4
1	2	5/4/05	34	127	151	9	9
1	3	5/4/05	4	8	28	4	22
2	1	5/4/05	45	86	40	7	0
2	2	5/4/05	17	91	103	25	5
2	3	5/4/05	3	23	27	14	17
1	1	5/11/05	37	97	94	0	17
1	2	5/11/05	31	171	145	12	4
1	3	5/11/05	10	3	22	9	16
2	1	5/11/05	51	81	77	10	6
2	2	5/11/05	22	65	109	22	1
2	3	5/11/05	34	38	61	26	15
1	1	5/18/05	23	123	116	13	21
1	2	5/18/05	44	76	87	8	7
1	3	5/18/05	5	0	34	0	28
2	1	5/18/05	37	47	74	14	4
2	2	5/18/05	52	68	98	17	0
2	3	5/18/05	12	47	17	21	9
1	1	5/26/05	45	81	83	4	6
1	2	5/26/05	27	98	113	11	0
1	3	5/26/05	9	1	16	7	18
2	1	5/26/05	39	98	69	21	2
2	2	5/26/05	33	79	86	20	2
2	3	5/26/05	3	57	31	7	11
1	1	6/1/05	23	103	62	3	0
1	2	6/1/05	8	191	91	2	0
1	3	6/1/05	46	42	48	8	0
2	1	6/1/05	9	196	18	1	0
2	2	6/1/05	13	137	165	0	1
2	3	6/1/05	3	44	24	2	5
1	1	6/10/05	20	168	42	7	0
1	2	6/10/05	19	179	62	3	0
1	3	6/10/05	27	17	73	4	0
2	1	6/10/05	24	174	43	0	1
2	2	6/10/05	16	151	87	0	1
2	3	6/10/05	6	23	51	1	1
1	1	6/24/05	42	139	86	1	0
1	2	6/24/05	16	187	71	9	0
1	3	6/24/05	34	11	96	18	0
2	1	6/24/05	16	245	38	1	3
2	2	6/24/05	11	67	91	0	0

Table 3.12. (cont.)

Station No	Depth[1]	Sampling Date	Chloro-phytae	Cyano-phytae	Pyrro-phytae	Eugleno-phytae	Chryso-phytae
2	3	6/24/05	1	17	74	0	3
1	1	6/30/05	25	150	58	5	0
1	2	6/30/05	3	195	56	6	0
1	3	6/30/05	39	18	59	10	0
2	1	6/30/05	22	217	26	2	0
2	2	6/30/05	7	113	149	0	2
2	3	6/30/05	5	20	43	1	3
1	1	7/8/05	36	161	78	12	3
1	2	7/8/05	27	139	64	16	4
1	3	7/8/05	21	18	51	26	9
2	1	7/8/05	21	109	63	8	1
2	2	7/8/05	30	89	87	6	0
2	3	7/8/05	38	93	46	10	7
1	1	7/15/05	27	70	81	20	10
1	2	7/15/05	36	87	101	27	0
1	3	7/15/05	16	41	37	9	7
2	1	7/15/05	21	21	76	50	3
2	2	7/15/05	10	91	103	3	0
2	3	7/15/05	6	18	31	1	7
1	1	7/30/05	31	152	90	9	0
1	2	7/30/05	23	119	76	4	7
1	3	7/30/05	14	35	80	3	3
2	1	7/30/05	40	117	121	10	2
2	2	7/30/05	37	198	107	3	0
2	3	7/30/05	10	97	162	7	4

Table 3-13. Relative composition (%) of the phytoplankton community structure at Guajataca during the study period (March 05 – July 05). Refer to Appendix F for complete statistical report.

Phytoplankton Division	Entrance	Dam	MSD[1]
Chlorophytae	12.26 a	9.12 b	1.829
Cyanophytae	43.22 a	38.47 a	4.806
Pyrrophytae	38.20 a	38.03 a	3.537
Euglenophytae	5.62 a	5.28 a	1.928
Chrysophytae	5.45 a	4.34 a	2.177

[1] – Minimum significant difference

Table 3-14. Effect of sampling depth on phytoplankton species richness (number of individuals) at Guajataca

	Sampling Depth			
Phytoplankton Division	**0M**	**1M**	**5M**	**MSD[1]**
Chlorophytae	25.70 a	23.94 a	12.11 b	5.89
Cyanophytae	113.33 a	119.89 a	26.44 b	19.55
Pyrrophytae	78.86 a	90.47 a	42.11 b	13.33
Euglenophytae	7.83 a	9.95 a	8.39 a	3.78
Chrysophytae	3.47 b	3.28 b	9.81 a	2.67

[1] – Minimum significant difference

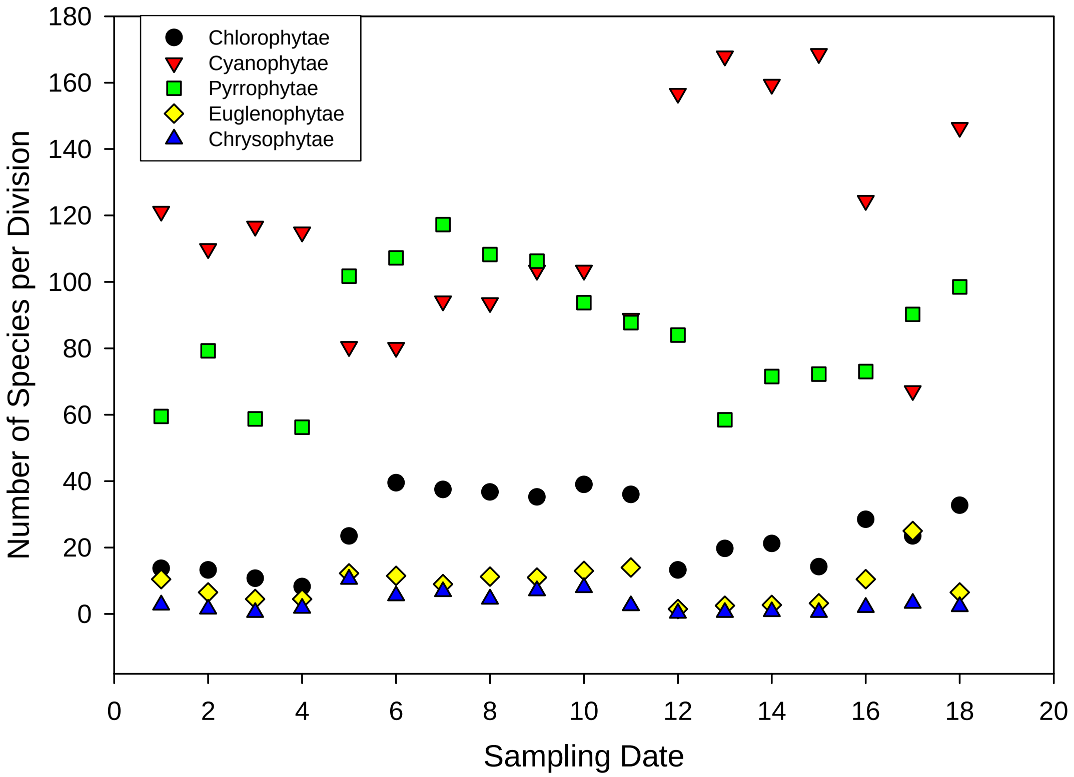

Figure 3.20: Changes in the average number of species of different phytoplankton divisions at Guajataca. Results exclude the 5m sampling depth.

4.0 PHYTOPLANKTON COMMUNITY OF THE PRINCIPAL RESERVOIRS OF PUERTO RICO

4.1. INTRODUCTION

Reservoirs constitute one of the main anthropogenic impacts on the hydrological regime of rivers (Baxter, 1977 cited by Bonilla and Martínez, 1999, Roldán, 1992). During the last decades, the demographic growth in Latin America and the Caribbean has accelerated the building of reservoirs for power generation and potable water supply. Since reservoirs are more susceptible to human impact, these systems require monitoring and evaluation. Thus, studies on the phytoplankton community are relevant to manage water quality.

The studies concerning taxonomic diversity of phytoplankton from reservoirs in Puerto Rico are limited. Jobin *et al*. (1979) realized the most complete survey available to date on the water chemistry and plankton abundance in our reservoirs. Martínez (1979) conducted a comparative study based on water quality for 10 major reservoirs. The results of these two studies demonstrated the advanced eutrophic state of the major reservoirs. The Puerto Rico Environmental Quality Board produced a preliminary trophic classification and priority ranking for the restoration of these reservoirs (PEQB, 1984).

In 1980 Quiñones-Márquez studied reservoir Carraízo, also known as Lake Loíza, the results were published in a comprehensive treatise for the U.S.G.S. This reservoir has also been studied by de Jesús (1979), Candelas (1981) and Candelas *et al*. (1992), PRASA (1992, 1995a-b), Montañez (1997), and Gellis *et al*. (1999). In the La Plata reservoir, García and Tilly (1982, 1983) conducted a one year study on the dynamics of phosphorus and nitrogen. In 1997, Ramos-Ginés produced a model for the total nitrogen and total phosphorus budgets of Lake Cidra. The rest of our reservoirs are under-investigated, and little is known about their plankton composition.

The aim of this study was to characterize the phytoplankton composition and abundance of reservoirs in Puerto Rico (Carite, Patillas, Guayo, Caonillas, Matrullas, Cerrillo, Cidra, Guineo, Dos Bocas, Melania, Loco, Luchetti, Garzas, Guayabal, Toa Vaca, Las Curías, Guajataca, Carraízo and La Plata).

4.2. SAMPLING AND ANALYSES

The present chapter is part of the overall scope of this study to relate the trophic status of lakes to biological diversity. Phytoplankton samples were taken at approximately quarterly intervals by the Environmental Quality Board (See Chapter 2). Sampling was performed between August 2003 and July 2005 in entrance, center and dam sections of each lake. Eighteen lakes were included in the study, and data for only thirteen lakes are reported. Sampling was classified *a posteriori* into sampling during “wet” and “dry” season (See Chapter 2). Phytoplankton samples were taken with plankton nets style Wisconsin of 48 μm mesh size. The collected samples were transferred to 500 ml bottles and fixed in glutaraldehyde to a concentration 2%. It was used the classification system of Drouet (1942, 1968, 1973, 1978, 1982) and Drouet and Daily (1939, 1952, 1956) to Class level.

Algal counts were made in Sedgwick – Rafter mounts until 50 fields and 300 identified cells had been counted. Identification of taxa was done with a light microscope (Nikon Optiphot) with magnifications of 1500x, and was followed by measurement of species populations.

4.3. RESULTS AND DISCUSSION

Certain algal groups were probably excluded from sampling because a 0.45 µm size mesh net was utilized. Pico-plankton (diameter 0.2 to 2 µm) are usually the dominant algal groups that contribute the greatest amounts of Chlorophyll a (Bonilla, pers. Comm.). We quantified micro-plankton (diameter 20 to 200 µm), so that small phytoflagellates and other algae of size less than 48 µm were not quantified. A total of 70 taxa (Table 1, Appendix G) can be considered as regular planktonic organisms in lakes of Puerto Rico. This is based on observations from more than 40 samples collected on different dates and locations within the lakes from various selected lakes. Of the 70 taxa identified, species richness was highest in the Chlorophyta (green algae) (34 taxa), which mainly comprised taxa of the orders Desmidiales and Chrorococcales. Bacillariophyta had 16 taxa, Dinophyta had 7 taxa, Cyanobacteria had 7 taxa, and Chrysophyta were less diversified.

Microcystis was the only algal group found in all reservoirs. According to Reynolds and Rogers (1976, cited by Akin-Oriola, 2003) who studied the distribution and buoyancy of *M. aeruginosa*, colonies stay over winter in a vegetative form on bottom sediments and return to the epilimnion later in spring or summer. The development of phytoplankton blooms in eutrophic lakes is attributed to their ability to thrive under conditions of low nitrogen to phosphorus ratios. With respect to other algae they are not predated as much as other algal groups due to the large colony sizes that they form. Other genera that were observed in high frequency in most lakes were the genera *Fragilaria*, *Gomphonema* and *Pediastrum*, which are indicators of eutrophic conditions, and usually appear under high N/P ratios.

Table 4.1. Presence of algal divisions, genera and species (when possible) in lakes of Puerto Rico.

Reservoir	**1**	**2**	**3**	**4**	**5**	**6**	**7**	**8**	**9**	**10**	**11**	**12**	**13**	**14**	**15**	**16**	**17**	**18**	**19**
CHLOROPHYTA																			
Gonatozygon sp.													1						
Ankistrodesmus sp.		1						1		1			1			1	1		1
Sp. 1 cf. Closterium	1	1	1			1	1							1			1	1	
Closterium sp.				1	1			1	1	1	1		1		1		1		1
Pandorina sp.			1		1	1	1					1					1	1	
Cosmarium sp. 1		1		1		1			1	1			1		1		1	1	1
Cosmarium sp. 2			1				1	1			1	1						1	
Pediastrum simplex var.1	1	1	1	1	1	1	1	1		1		1	1	1		1	1	1	1
Pediastrum simplex var.2	1		1			1		1				1	1	1					
Pediastrum simplex. var.3		1			1	1	1	1				1	1					1	
Pediastrum duplex						1										1		1	1
Staurastrum sp. 1		1	1		1	1			1		1		1		1		1	1	1
Staurastrum sp. 2			1	1	1	1	1					1		1			1	1	
Staurastrum sp. 3	1						1	1		1									
Micractinium sp.			1		1	1	1					1						1	
Tetraedron	1	1	1		1	1	1	1		1		1	1			1	1		1
*Secenedesmus sp.*1	1	1	1			1	1		1	1			1			1	1		1
Scenedesmus sp.2	1				1	1	1	1				1						1	
Scenedesmus sp.3					1		1												
Oocystis sp.	1	1	1	1	1	1		1		1		1	1				1	1	
Monoraphidium sp.		1	1	1	1	1	1	1		1			1	1			1	1	1
Eudorina sp.						1							1					1	
*Ankistrodesmus sp.*2			1									1							
Dictyosphaerium sp. 1		1	1	1			1												
Dictyosphaerium sp. 2												1							
Selenastrum sp.												1							
Euastrum sp.		1		1				1		1	1		1		1		1	1	1
Asterococcus sp.												1		1					
Xanthidium sp.	1	1		1				1		1		1			1		1		1
Botryococcus sp.				1				1			1			1				1	
Treubaria sp.		1		1			1			1			1						
Coelastrum sp.		1		1	1			1		1	1		1			1	1	1	1
Spirogyra sp.		1		1				1	1	1	1		1		1		1	1	1
Sp. 5 cf. Crucigenia		1	1	1		1		1	1	1	1		1			1	1	1	1
BACILLARIOPHYTA																			
Fragilaria sp.1	1	1		1	1	1	1	1	1	1	1	1	1				1	1	
Fragilaria sp.2													1		1	1			1
Synedra sp. 1		1	1	1	1	1	1					1	1	1			1	1	
Synedra sp. 2			1		1	1												1	
Sp.9 cf. Navicula						1	1				1	1						1	
Navicula sp. 1	1		1	1	1	1						1							
Amphora sp.												1							
Gomphonema sp.	1	1		1	1		1	1	1	1	1		1	1		1	1	1	1
Navicula sp. 2					1									1					
Amphiprora sp.		1		1			1	1	1	1	1		1			1		1	1

Table 4.1 (cont.)

Reservoir	**1**	**2**	**3**	**4**	**5**	**6**	**7**	**8**	**9**	**10**	**11**	**12**	**13**	**14**	**15**	**16**	**17**	**18**	**19**
Cyclotela sp.	1	1		1	1		1	1	1	1	1		1				1		1
Nitzchia sp.				1	1		1				1		1					1	
Surirella sp.	1				1		1											1	
Cymbella sp.	1				1			1	1	1	1						1	1	1
Gyrosigma sp.		1			1													1	
Cocconeis sp.					1														
CHRYSOPHYTA																			
Dinobryon sp.		1		1	1		1				1	1							1
DINOPHYTA																			
*Peridiniopsis sp.*1	1	1	1		1	1	1					1	1				1		
*Peridiniopsis sp.*2											1						1		
Peridinium sp. 1	1	1	1		1	1	1						1	1					
Peridinium sp. 2	1	1	1	1							1	1	1	1			1		
Peridinium sp. 3					1														
Ceratium sp																			
cf. Protoperidinium													1						
EUGLENOPHYTA																			
Phacus sp.	1		1	1		1				1				1					1
Euglena sp.1		1	1	1	1	1		1	1	1			1	1	1	1	1	1	1
Lepocinclis sp.			1			1	1				1	1						1	
Strombomonas sp.			1																
Trachelomonas sp.	1	1	1		1			1	1	1		1	1	1	1	1	1		1
CYANOBACTERIA																			
Microcystis sp	1	1	1	1	1	1	1	1	1	1	1	1	1	1	1	1	1	1	1
Sp.5cf. Synechocystis			1			1						1							
Cylindrospermopsis sp		1	1	1					1	1		1							
Chroococcus sp.	1	1		1	1		1	1		1	1	1	1	1	1	1	1	1	
Oscillatoria sp.				1	1		1	1	1	1	1	1		1	1			1	1
Gomphosphaeria sp.	1	1			1			1		1	1	1	1		1	1	1	1	
Merismopedia sp.	1	1			1				1		1	1	1		1	1	1		1

1= Curias, 2=La Plata, 3= Carraízo, 4= Carite, 5= Patillas, 6= Guayo, 7=Caonillas, 8= Matrullas, 9= Cerrillo, 10= Cidra, 11= Guineo, 12= Guajataca, 13= Dos Bocas, 14= Melania, 15= Loco, 16= Luchetti, 17= Garzas, 18= Guayabal, 19= Toa Vaca

In terms of abundance, the dominant species were those belonging to the genera *Pediastrum, Peridinium* and *Botryococccus*. *Pediastrum* was dominant for the lakes Guayo, Guajataca, Melania and Curias. *Peridinium* was dominant for the lakes Carraízo, Guayabal and Patillas, while *Botryococcus* was dominant in Carite. Algae of the genus *Pediastrum* are ones that stay at the surface and are associated with "off-fish" flavor of the water. *Pediastrum, Peridinium* and *Botryococccus* are indicators of eutrophic conditions, yet these in this study, these were found in both types of lakes, i.e. those that are in the eutrophic classification and those in the meso- and hypotrophic classifications (based on TSI(TP)). This may occur study due to low predation by zooplankton. Another explanation is that with the use of larger size net, *Pediastrum, Peridinium* and *Botryococccus* were selected for, and the smaller size algal groups were excluded. Further work is needed to ascertain this hypothesis.

The species *P. simplex* is associated with areas that receive wastes rich in salts (Pinilla, 2000), especially sulfates and sodium chloride. *Botryococcus* are planktonic algae, which tend to appear in greater number in lakes used for public water supply (Branco, 1986) and eutrophic conditions. *Perinidium* is the dinoflagellate genus most abundant in freshwaters (Prescott, 1970). It is an indicator of eutrophic conditions and usually share dominance with cyanobacteria.

Overall, the mean relative proportional composition (%) of Chlorophyta, Ochrophytes, Dinophyta, Cyanobacteria, and Euglenophyta in all lakes is shown Figure 4.1. The distribution followed the order: Chlorophyta > Dinophyta > Ochrophytes > Cyanobacteria > Euglenophyta.

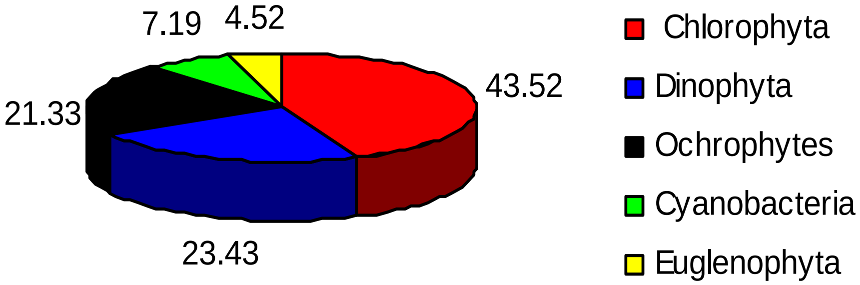

Figure 4.1. Percentage composition of phytoplankton in 13 reservoirs of Puerto Rico

The division Chlorophyta had the higher representation in all samplings. Within the Chlorophytas, the non-desmidiaceas were most abundant, with the latter being indicators of eutrophic conditions and high N/P ratios. In addition, the genus *Pediastrum*, *Botryococcus*, *Cosmarium*, *Closterium*, of the Chlorophyta division are indicators of eutrophic environments. Chlorophyceas are known as sun algae (Roldán, 1992), that is, adapted to high illumination conditions. The fact that Chlorophytas were found in highest numbers, and that the types of alga found within Chlorophyta division are associated with eutrophic conditions, suggests that there is a high probability for present and potential water-quality degradation of lakes of Puerto Rico.

The division Dinophyta, represented by *Peridinium*, *Perdiniopsis,* and *Ceratium*, obtained the second position in abundance. *Peridinium* was more abundant during dry period than in the rainy periods. For Marquez and Guillot (1988) and Wetzel (1989) the genus *Peridinium* is present in lakes with low in nutrients, neutral or lightly alkaline and Ca rich waters. However, in this study *Peridinium* was present in lakes with varying trophic state of both hypotrophic, mesotrophic and eutrophic conditions.

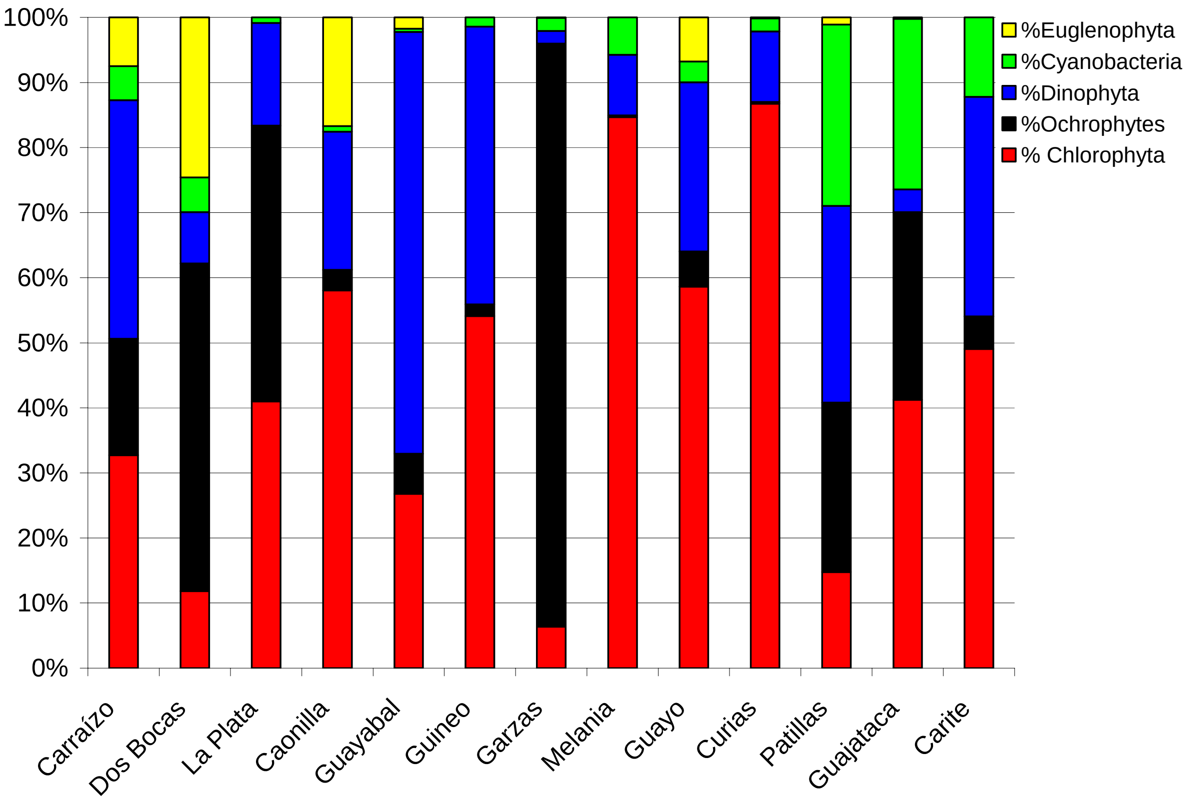

Figure 4.2. Percent Composition Phytoplankton of the 13 Lakes in Puerto Rico

The division Ochrophyta was third in abundance. Within this division were the classes Chrysophyceaes and Bacillariophycaes. The genera *Synedra* and *Fragilaria* were more abundant in Lago Dos Bocas and La Plata, while *Dinobryon* was present in Lago Garzas (Figure 2). The bacillariophyceas are associated with neutral pH, eutrophia, high N/P ratios and littoral waters of lakes. We observed that these genera were most abundance in the entrance of the lakes (riverine stations). The genus *Dinobryon* has been associated with oligotrophic conditions. However, with high abundance of this algae, the lake water quality could be affected because it imparts a cucumber-like smell to water.

The presence of algae of the Cyanobacteria division is characteristic of environments ranging from eutrophic to hypereutrophic conditions (Duque and Donato, 1992) and have been found to proliferate under N limiting conditions, due to their ability to fix N. Cyanobacteria were in low abundance in all lakes evaluated except for Lago Carite, Patillas, and Guajataca. Of the Cyanobacteria, *Microcystis*, *Chroococcus* and *Oscillatoria* were most abundant.

The algae of the division Euglenophyta had a lower representation in this study, but was of importance in Carraízo, Dos Bocas and Caonillas, which has high P and N concentrations (Figure 2). The genera most representative were *Euglena*, *Trachelomonas*, *Lepocinclis* and *Phacus*. The presence of euglenophyceaes in these lakes could be attributed to high organic matter inputs and organic matter in lakes. These algae have the capacity to utilize both organic and inorganic carbon as energy sources (facultative heterotrophy).

Thirty-six of the forty sampling events evaluated were sampled during a time period classified as “dry”. In Dos Bocas, there was a shift in Ochrophytes, Dinophyta, Cyanobacteria, from 37 to 63%, 0 to 15%, and 0 to 11%, from wet to dry periods respectively. In Euglenophyta the relative abundance changed from 49% during the wet period to none in the dry period. Further analyses need be done to corroborate possible shifts in algal abundance in wet versus dry periods.

The algal diversity range for the lakes in this study was between 0.27 bits for Garzas and 2.78 bits for Guayo. Mean diversity values for all lakes was 1.5 bits (standard deviation = 0.7). Diversity values above 3.0 bits is considered a diverse system. In general, the diversity levels are indicative of meso- to eutrophic conditions, where few species are more abundant. This information further reinforces was has been previously been observed using the trophic state index approach.

There was considerable algal spatial heterogeneity in phytoplankton assemblages in the data observed in this study. That is, we observed high variation in algal composition between the riverine and lacustrine portions of lakes. Establishment of the principal factors that affect the composition and distribution of the phytoplankton in tropical lakes and reservoirs is not an easy task (Albano and Matsumura-Tundisi, 1997). Most lakes showed high phytoplankton similarities in algal composition of groups such as Chlorococcaceae and Desmidiaceae. This may be explained by the input of nutrients and dissolved organic matter due to runoff from the watershed and riverine input. Other factors such as the water flow in the direction of the day, and increased organic matter decomposition could be important controls of the phytoplankton assemblages. Although phytoplankton species composition and diversity changes with environmental conditions such as nutrient levels, temperature, light, predator pressure, etc., the relative importance of these factors varies considerably among taxa (Akin-Oriola, 2003). Under conditions of enrichment or eutrophication, the cyanobacteria are known to proliferate and form noxious blooms in freshwaters environments.

The lakes were grouped according to their mean TP versus TKN/TP ratios, demonstrating a clear negative exponential relationship (Figure 4.3). Other relationships evaluated (TP versus TN/TP; TN versus TN/TP; and TKN versus TKN/TP) showed greater scatter and were clearly non-significant. The lakes associated with eutrophy and hypereutrophy, namely Lago Carraizo, Dos Bocas and La Plata clearly had lower TKN/TP ratios, whereas the lakes associated with hypotrophy and mesotrophy (Carite, Patillas, Guajataca and Curias) had greater TKN/TP ratios and lower TP concentrations. The results demonstrate a clear N limitation in eutrophic and hypereutrophic lakes and a clear P limitation in hypotrophic and some mesotrophic lakes.

When the reservoirs were grouped according to their TKN/TP ratios, distinct shifts in algal predominance are observed. Cyanobacteria were present in a considerable proportion in lakes with high TKN/TP ratios while Euglenophyta were mostly present in lakes with low TKN/TP ratios, with a percentage composition of 22.8 vs. 3.8 % (low vs high TKN/TP) for Cyanobacteria and 0.4 vs. 10.7 % (low vs high TKN/TP) for Euglenophyta. Algae of the Ochrophyta division have competitive advantage at low P concentrations which may explain the high relative proportion of this division in Lago Garza which has average TP concentrations of 0.021 mg P/L. The presence of cyanobacteria in lakes with high TKN/TP ratios is clearly justified because there is very low N available, which favors the presence of N fixing algae. The presence of Euglenophyta in lakes with high TKN and TP concentrations, but low TKN/TP ratios, is explained by probable inputs of dissolved organic matter and nutrients. In fact, Peridium spp. were most abundant in Lago Carraizo.

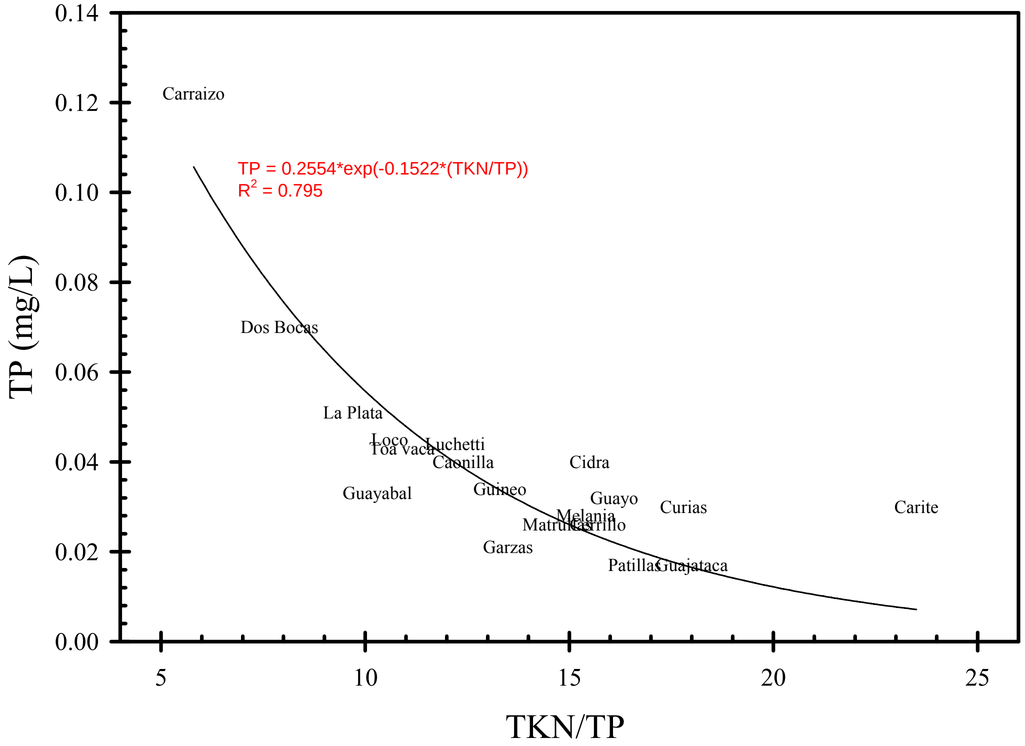

Figure 4.3. Relationship between average TKN/TP ratios and average TP concentrations for reservoirs of Puerto Rico

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6.0 ADMINISTRATIVE

Professional hours devoted to project

Table 6.1: Professional hours devoted to project (by activities).

Tasks	Professional hours	Total hours
Simple pick up at EQBs laboratory	8 hrs/ sampling event	64
Filtering of simples for DP, and DOC determination	8 hrs/ sampling event	64
Digestion – TKN	16 hrs/ sampling event	128
Digestion –TP	12 hrs/ sampling event	96
Cleaning glassware	40 hrs/ sampling event	320
DOC analysis	3 hrs/ sampling event	24
TP, DP analysis	8 hrs/ sampling event	64
TKN analysis	8 hrs/ sampling event	64
Preparation of laboratory Report	16 hrs/ sampling event	128
Chlorophyll analysis - Extraction, and sample measurement	24 hrs/ sampling event	192
Data evaluation- (e.g., preparation of graphs, data manipulation)	24 hrs/ sampling event	192
Preparation of purchase orders for materials, and supplies	58 hrs	58
Financial report preparation	24 hrs	24
Statistical Analyses	80 hrs	80
Preparation of Progress Reports	200 hrs	200
Total hours		**1698**

Table 6.2. Professional hours devoted to peripheral tasks related to the project

Other tasks related to the project	Professional hours
Development of electronic data base	288
Periphytometer construction	48
Periphytometer studies (15 trials @ 144 hrs/trial (3 persons @ 48 hrs each/trial)	2,160
Chemical and Biological analyzes (TKN, TP, DOC, Chl *a*.) –Guajataca Monitoring study	100
Phytoplankton characterization –Guajataca Monitoring Study study (576 slides (96 samples-6 slides per sample) @ 2 hours per slide)	1,152
Total	**3,748**

Table 6.3. Professional hours devoted in the phytoplankton characterization study:

Number of lakes	Average Number of samples per lake	Number of visits	Number of slides scanned per sample	Time /slide (including species identification)
19	**4**	**4**	**6**	**1.5-2 hrs.**
			TIME EFFORT	**594-1296 hours (25-54 straight days)**

7.0 APPENDIX

Table 7.1 Appendix List

Identification	Description
Appendix A	Chemical and Biological Characterization of Reservoir Samples
Appendix B	Statistical analyses of reservoir data
Appendix C	Chemical and Biological Characterization of samples from the Guajataca Monitoring study
Appendix D	Statistical analyses of samples from the Guajataca Monitoring study
Appendix E	Characterization of Phytoplankton community structure on samples from the Guajataca Monitoring study
Appendix F	Statistical analyses of phytoplankton data from the Guajataca Monitoring study
Appendix G	Characterization of Phytoplankton community structure on reservoirs of Puerto Rico