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Summary of nutrient enrichment studies of natural phytoplankton populations in the Lower Hillsborough River, the Palm River, and the Alafia River conducted on October 18 - 22, 2004

The City of Tampa Wastewater Department

SUMMARY OF NUTRIENT ENRICHMENT STUDIES OF NATURAL PHYTOPLANKTON POPULATIONS IN THE LOWER HILLSBOROUGH RIVER, THE PALM RIVER, AND THE ALAFIA RIVER CONDUCTED ON OCTOBER 18 - 22, 2004

THE CITY OF TAMPA
WASTEWATER DEPARTMENT
BAY STUDY GROUP

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INTRODUCTION

The Bay Study Group (BSG) conducted nutrient enrichment studies (bioassays) on natural phytoplankton population samples collected in the Lower Hillsborough River, the Palm River, and the Alafia River on October 18, 2004. The Lower Hillsborough River test site was located approximately 200m upriver of the Nebraska Avenue bridge (Lat: 28^{o} 01.171'N; Lon: 82^{o} 27.025'W), the Palm River test site was located approximately 300m downstream from the barrier at Structure-160 (Lat: 27^{o} 57.308'N; Lon: 82^{o} 22.200'W), and the Alafia River test site was located at the Hillsborough County boat ramp at Center Avenue (Lat: 27^{o} 52.842'N; Lon: 82^{o} 18.011'W).

METHODS

The bioassays were performed on the natural phytoplankton populations collected from surface waters of the three test sites. All water samples were collected from a small boat near the center of each stream.

The bioassay method used was similar to a method that has been used in Tampa Bay and Chesapeake Bay waters (see City of Tampa 1992; Fisher et al. 1992a and 1992b). A summary of the method used by the BSG is provided here.

A large volume of surface water was used to provide the following nutrient treatments. Each treatment was conducted in duplicate on 3l samples:

- Controls (no nutrient addition).
- Nitrogen (N) additions (NH3-N added to reach the final treatment concentrations shown in Table 3.
- Phosphorous (P) additions (PO4-P added to reach a the final treatment concentrations shown in Table 3.
- N+P-additions (combination of the respective N-additions and P-additions).

The treatment samples were incubated outside under natural sunlight (incident radiation reduced by approximately 40 percent from a neutral density screen) in a water-cooled deck incubator. The incubation periods for the Lower Hillsborough River, the Palm River, and the Alafia River were 72hrs, 69hrs, and 91hrs, respectively.

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

Paired t-test statistics were used to interpret the bioassay results and to classify the growth response to the nutrient additions into the following response categories:

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

RESULTS

Water quality conditions and field observations at the three river sampling locations on October 18, 2004 are shown in Table 1.

Table 1. Ambient water quality conditions and observations at the three river sample locations on October 18, 2004.

Parameter	**Station**		
	Hillsborough River	**Palm River**	**Alafia River**
Time (hhmm)	0829	1116	1004
Surface temp. (C)	23.0	26.4	21.5
Surface DO (mg/l)	5.98	4.11	6.04
Surface salinity (PSU)	0.1	13.1	0.23
pH	7.19	7.19	7.47
Secchi depth (m)	0.7	1.0	1.2
Water column depth (m)	3.7	5.6	5.3
Apparent water color	Brown	Brown	Brown
Flow and turbulence	Yes	Yes	Yes
Turbidity (NTU)	1.0	1.8	4.2
Chlorophyll-a (ug/l)	3.75	5.64	1.26

Ambient surface nutrient concentrations at the three river sample locations are shown in Table 2. The Environmental Protection Commission of Hillsborough County kindly provided the analyses.

Table 2. Ambient surface nutrient concentrations at the three river sample locations on October 18, 2004.

Parameter (uM)	Station		
	Hillsborough River	Palm River	Alafia River
NH3	7.9	12.1	17.9
TKN	85.7	62.9	111
NO3+NO2	6.8	8.4	92.1
TN	92.9	71.4	203
PO4	8.4	13.2	54.5
TP	9.4	13.5	71.9
SiO2	246	157	290

The nitrogen (NH3) and phosphorous (PO4) additions to the bioassay treatment sample containers are shown in Table 3. The table also shows the final nutrient concentration in the treatments, which includes the nutrient addition plus the ambient nutrient concentrations.

The Hillsborough River sample will be used as an example to further illustrate Table 3 and the four bioassay treatment combinations: (1) Duplicate control sample containers received no nutrient additions and these containers only contained the ambient nutrient concentrations shown in Table 2. (2) Duplicate containers received the NH3 treatment (23.9uM) in addition to the ambient nutrient concentrations. (3) Duplicate containers received the PO4 treatment (2.9uM) in addition to the ambient nutrient concentrations. (4) Duplicate containers received both the NH3 treatment (23.9uM) and the PO4 treatment (2.9uM) in addition to the ambient nutrient concentrations.

Table 3. Concentrations of treatment nutrient additions to the three river surface samples and the final concentrations in respective treatments prior to incubation on October 18, 2004.

Parameter (uM)	Station		
	Hillsborough River	Palm River	Alafia River
NH3 treatment addition	23.9	23.9	23.9
NH3 final treatment conc.	31.8	36.1	41.8
PO4 treatment addition	2.9	2.9	38.4*
PO4 final treatment conc.	11.3	16.1	92.9

* The Alafia River sample required a higher addition of PO4, than the other river samples, in order to obtain a substantial final treatment concentration of PO4 above the expected, and later confirmed, high ambient PO4 concentration.

Table 4. Dissolved inorganic nitrogen (DIN) and reactive phosphorous (SPR) ratios for the ambient samples, the nutrient additions and the final treatments prior to incubation on October 18, 2004.

Station	Ambient	Additions	Final treatments
Hillsborough River	1.8	8.2	3.4
Palm River	1.6	8.2	2.8
Alafia River	2.0	0.6	1.4

Phytoplankton biomass (chlorophyll-a) was measured in the bioassay containers following the incubation periods shown in Table 5. Differences in biomass in the nutrient addition treatment containers relative the control containers were analyzed using t-test statistics. The results from these analyses were grouped into the biomass response categories described above. Further, the biomass response to the nutrient treatments at each of the three river locations is summarized in Table 5.

Table 5. Results from natural phytoplankton nutrient bioassays on surface waters from the three rivers. Samples were collected, and the bioassays were initiated, on October 18, 2004.

Station	**Incubation time (hr)**	**T-test results of nutrient addition treatments vs. controls**			**Response**
		NH3 vs. control	**PO4 vs. control**	**NH3+PO4 vs. control**	
Hillsborough River	72	**	NS	**	Exclusive nitrogen limitation
Palm River	69	**	* (control>PO4)	**	Exclusive nitrogen limitation
Alafia River	91	NS	NS	**	Balanced

** Significant difference at $p<0.01$
* Significant difference at $p<0.05$
NS = no significant difference
All tests 5df

DISCUSSION AND CONCLUSION

At the termination of the bioassay experiments, following the incubation times shown in Table 4, chlorophyll-a analyses indicated that the phytoplankton populations of the Lower Hillsborough River and the Palm River were “exclusively” limited by nitrogen (as defined by the response categories listed above). Additions of phosphate alone to these populations did not increase biomass significantly above the control treatments. Nor did the combined additions of nitrogen (ammonia) and phosphate increase biomass significantly above the nitrogen alone treatments. For the Alafia River sample, phytoplankton biomass did not increase statistically above the control treatment from additions of nitrogen alone, or phosphate alone. However, the combined nitrogen and phosphate treatment caused a highly significant biomass increase, resulting in a “balanced” response (as defined by the response categories listed above) to the nutrient additions. This response suggests that both nitrogen and phosphate limited phytoplankton growth.

Results from these bioassay experiments should, however, be interpreted with caution when attempting to describe the actual response to potential additions of nutrients above ambient

concentrations to the sampled areas of the three river systems discussed herein. The incubation period of the experiments, which usually ranges from 24 to 48hrs for estuarine Tampa Bay samples, had to be extended by several days in the river experiments to achieve significant changes in biomass between nutrient addition treatments and the controls. The long incubation times may, therefore, have "forced" the treatments to indicate a response to nutrient additions that may not have occurred from similar nutrient additions to the natural river systems. The need for the extended incubation periods suggests that nutrient limitation may not have been the primary limiting factor to phytoplankton growth at the time of sampling in these specific areas.

The experimental phytoplankton populations were contained in sample vessels and the populations received levels of solar radiation during the experiments that should not have limited growth during most of the daylight period. In contrast, substantial turbulence was observed in the river systems that could be expected to disperse the phytoplankton throughout the relatively deep and highly colored water column of the sampled areas (depth ranged from 3.7 to 5.6m). As a result, the phytoplankton populations would not always be near the surface at these sites and would thus receive substantially lower levels of solar radiation in comparison to the experimental populations. It is therefore likely that the ambient phytoplankton populations, present in the areas at the time of sampling, were primarily light limited.

The following supports the theory that the ambient phytoplankton populations may have been primarily light limited. The ambient biomass concentrations at the three locations were relatively low, however, once the phytoplankton populations were enclosed in the treatment containers, biomass generally increased equally rapid in both the control and nutrient addition treatments. The biomass increase that occurred over several days in all treatments, including the control, suggests that the ambient river waters contained a sufficient amount of nutrients to sustain a substantial and prolonged increase in biomass above the biomass present at the time of sampling. It was not until later in the experiments, when the biomass in all treatment containers (including the control) had increased substantially above the initially measured ambient river biomass concentrations, that nutrient limitation became evident.

The results from these tests suggest that the river phytoplankton populations present when the samples were collected may have been primarily light limited and that a potential nutrient addition to these areas, at that time, may not have caused an immediate (within 48hrs) increase in phytoplankton biomass. However, it should be recognized that potential nutrient additions above ambient concentrations to these river systems might result in increased phytoplankton biomass downstream and/or ultimately in Tampa Bay.

REFERENCES

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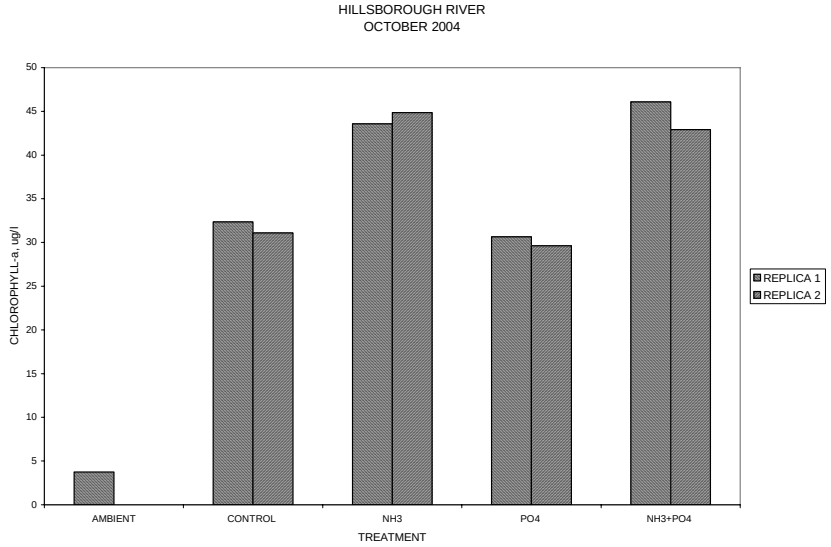
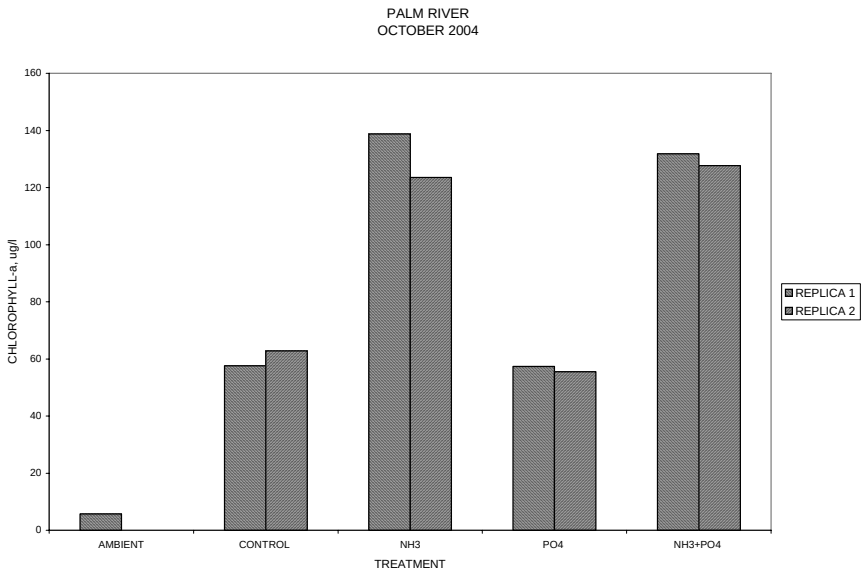
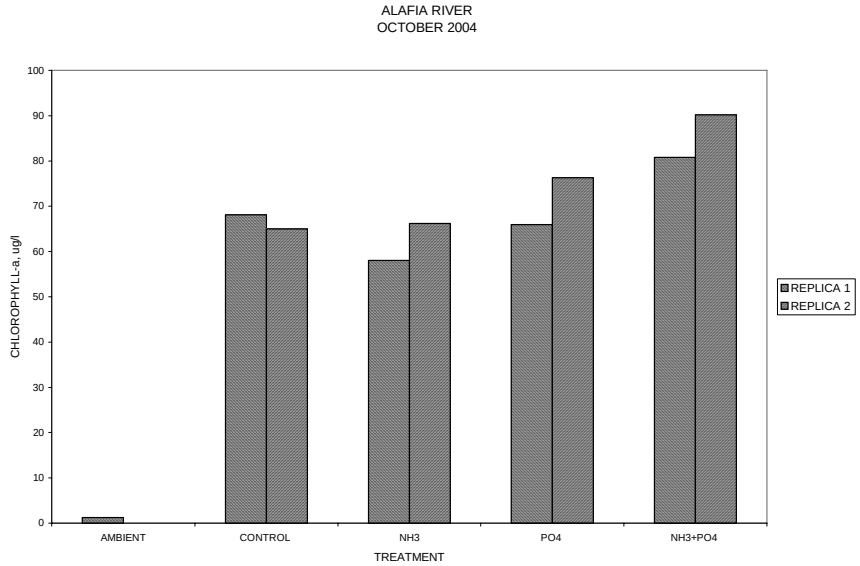
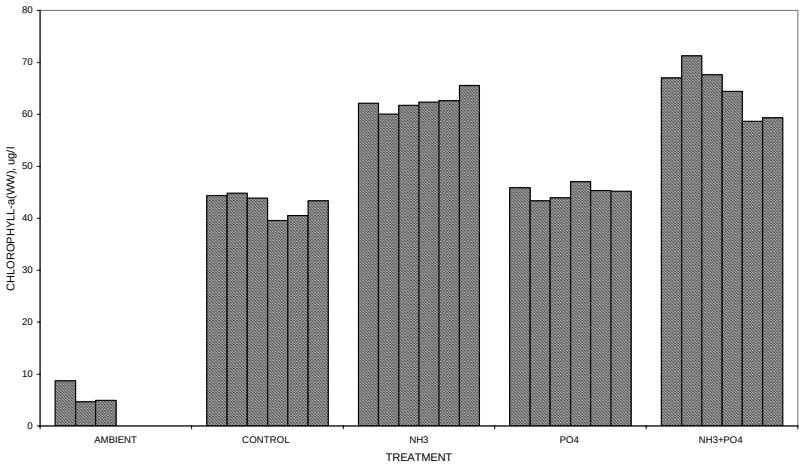




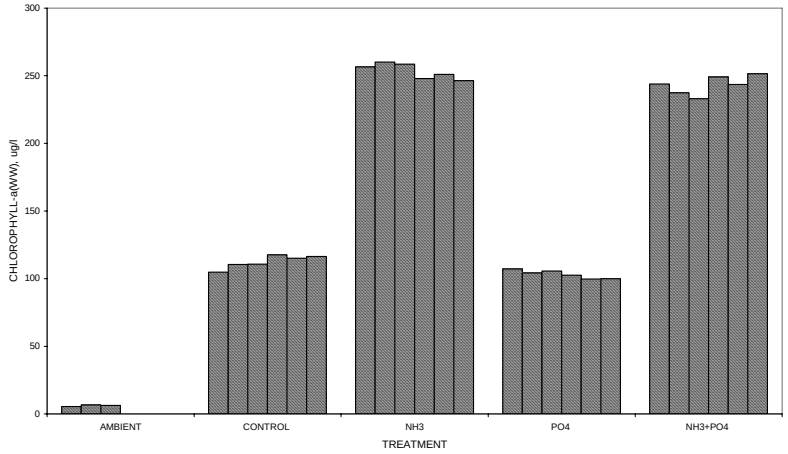


FLUOROMETRIC WHOLEWATER CHLOROPHYLL-A

HILLSBOROUGH RIVER
OCTOBER 2004



PALM RIVER
OCTOBER 2004



ALAFIA RIVER
OCTOBER 2004