

## **RESPONSE OF FOOD ORGANISMS TO INORGANIC NITROGEN AVAILABILITY.**

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(Received 24 July 2003; Revision accepted 21 November 2003)

### **ABSTRACT**

Influence of inorganic N<sub>2</sub> forms on pond food organisms was investigated. Seven identified plankton taxa comprising four phytoplankton: Desmidiaceae (desmids), Bacillariophyceae (diatoms), Cyanophyceae (blue-green algae) and Chlorophyceae (green algae) and three zooplankton: Protozoa, Cladocera and Rotifera fluctuated in response to nutrient inputs. All recorded significant F-value across treatments. With exception of blue-green algae, other phytoplankton groups had comparable relative abundances in treatment with higher concentration of readily utilized Nitrate-Nitrogen (NO<sub>3</sub>-N) compared with control that recorded higher concentration of Ammonia-Nitrogen (NH<sub>3</sub>-N) and increased relative abundance of blue-green algae. Zooplankton exhibited same dominance pattern of Protozoa, Rotifera and Cladocera respectively, with treatment recording higher abundance in response to edible phytoplankton. N<sub>2</sub> from zooplankton excretion influenced phytoplankton responses in the control. Ammonia-Nitrogen (NH<sub>3</sub>-N), Nitrate-Nitrogen (NO<sub>3</sub>-N) and Nitrite-Nitrogen (NO<sub>2</sub>-N) were observed to fluctuate along nutrient enrichment profile depending mostly on the concentrations of intermediate Nitrite-Nitrogen (NO<sub>2</sub>-N). All nutrient parameters except NH<sub>3</sub>-N recorded significant t and F statistic between and across treatments. Water quality parameter of pH and dissolved oxygen were favourable for most period of the experiment.

**KEYWORDS:** Nutrient, Plankton, Ammonia-Nitrogen, Nitrate-Nitrogen, Nitrite-Nitrogen.

### **INTRODUCTION**

Food organisms for pond culture referred to as plankton are limited by two main nutrients: phosphorus and nitrogen related to stoichiometric ratios (Hecky and Kilham, 1988). The importance of different N<sub>2</sub> forms is that most edible phytoplankton are not N<sub>2</sub> fixers and do not assimilate from diverse sources, but absorb inorganic forms (nitrates, nitrites and ammonia) present in water column (Falkowski and Raven, 1997). Therefore easily outnumbered and out-competed by fixers which are of low nutritional importance in pond culture (Pearl and Tucker, 1995) at periods of N<sub>2</sub> restriction. The results of this study aims to add to body of information on relationship between food organisms and different inorganic nitrogen forms.

### **MATERIAL AND METHODS**

The experiments were conducted at the Zoology Department, University of Ibadan, Nigeria, in 0.90m<sup>2</sup> partially shaded open concrete tanks, containing 2cm thick layer of sand. Water from Oba dam was used and maintained at 50cm mark. There were 2 sets of experiments each consisted of duplicated treatment and control. The experimental setup was maintained without fertilization for 1week to allow stabilization of

plankton population. Ammonium sulphate (0.01kg) was applied at the rate of 112kg/ha as recommended by Gratzek and Brown (1980). Two applications were made at 15 days interval. Thirty three days inorganic nitrogen profile was evaluated at 3-day interval through the concentrations of the following: Ammonia-Nitrogen (NH<sub>3</sub>-N), Nitrate-Nitrogen (NO<sub>3</sub>-N) and Nitrite-Nitrogen (NO<sub>2</sub>-N) measured respectively by phenate, phenol disulphonic acid and sulphanilamide methods recommended by Boyd (1979) and APHA (1985). Dissolved Oxygen Concentrations (DO<sub>2</sub>) was determined by Winkler's (Azide) titrimetric method (APHA, 1985) and pH by using PS17 Corning probe. Average water temperature for the duration of the experiment was 28.7°C. Plankton samples were collected simultaneously using 64µm bolting silk

net, and preserved with 4% formalin. The counting was done on a 15ml counting plate and composition determined by direct proportion in 1ml. Identification was with guides of Needham and Needham (1978), Pennak (1978) and Jeje and Fernando (1986). Treatment means were compared using t-test and One-way ANOVA (Sokal and Rohlf, 1987).

### **RESULTS**

The summary of experimental results is

Table 1. Summary and comparison of water quality parameters

	Treatments	Mean	t	F
Ammonia-Nitrogen (mg/l)	1	6.21±3.87		
	Control	4.66±1.61	1.65	
	2	6.42±4.13		1.20
Nitrate-Nitrogen (mg/l)	Control	4.67±1.37	1.72	
	1	5.87±5.48		
	2	4.99±5.47		3.59*
Nitrite -Nitrogen (mg/l)	Control	1.43±1.32	2.40*	
	1	5.69±2.5		
	2	1.79±1.40		20.81*
Dissolved oxygen (mg/l)	Control	0.32±0.12	3.39*	
	1	1.66±1.21		
	2	1.50±1.64		0.06
pH	Control	1.45±1.30	-1.21	
	1	7.28±0.45		
	2	7.19±0.44	0.44	
	Control	7.21±0.44		0.11
	2	7.28±0.47	0.36	

\* significant at 0.05

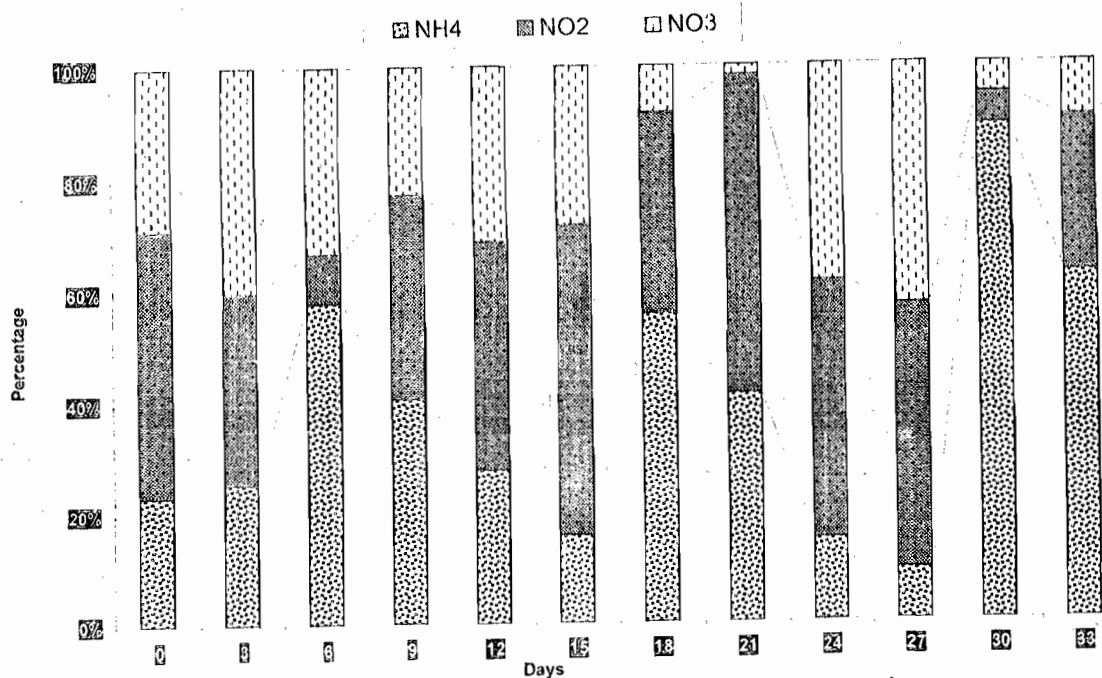


Fig.1 Percentage composition of different nitrogen forms in treatment tanks

presented in Table 1. Three  $N_2$  forms recorded higher mean treatment values in all cases compared with control.  $NO_3-N$  and  $NO_2-N$  recorded significant t and F values indicating significant mean differences between duplicates and across treatments. Changes in percentage proportion of  $N_2$  forms is shown in Figs. 1 & 2 for treatments and controls respectively.  $NH_3-N$  and  $NO_2-N$  exhibited inverse changes in concentrations, clearly depicted 6 days after first

application with noticeable increase in  $NH_3-N$  concentration followed by same magnitude of decrease till second fertilizer application. Similar post application changes were however observed after 3 days (Fig. 1) followed by near absence of  $NO_3-N$  and  $NO_2-N$ , with ~90%  $NH_3-N$ , which reduced to 65% at termination. The concentration of  $NO_3-N$  exhibited different profile with a 29% proportion at the commencement, which increased to about 46% and decreased

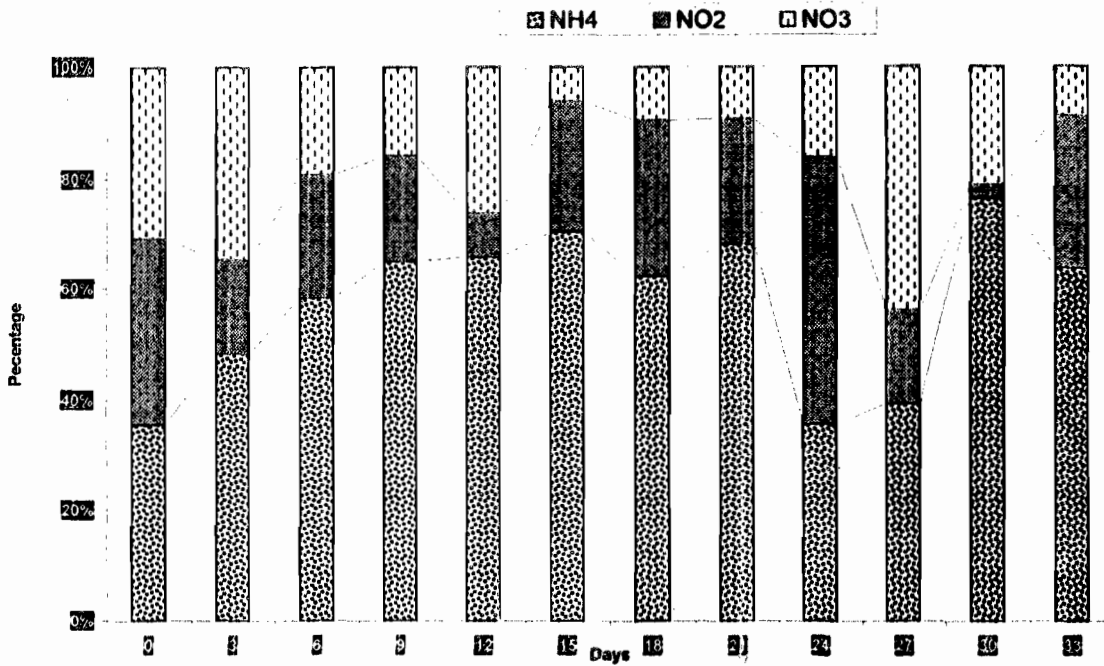


Fig.2 Percentage composition of nitrogen forms in control tanks

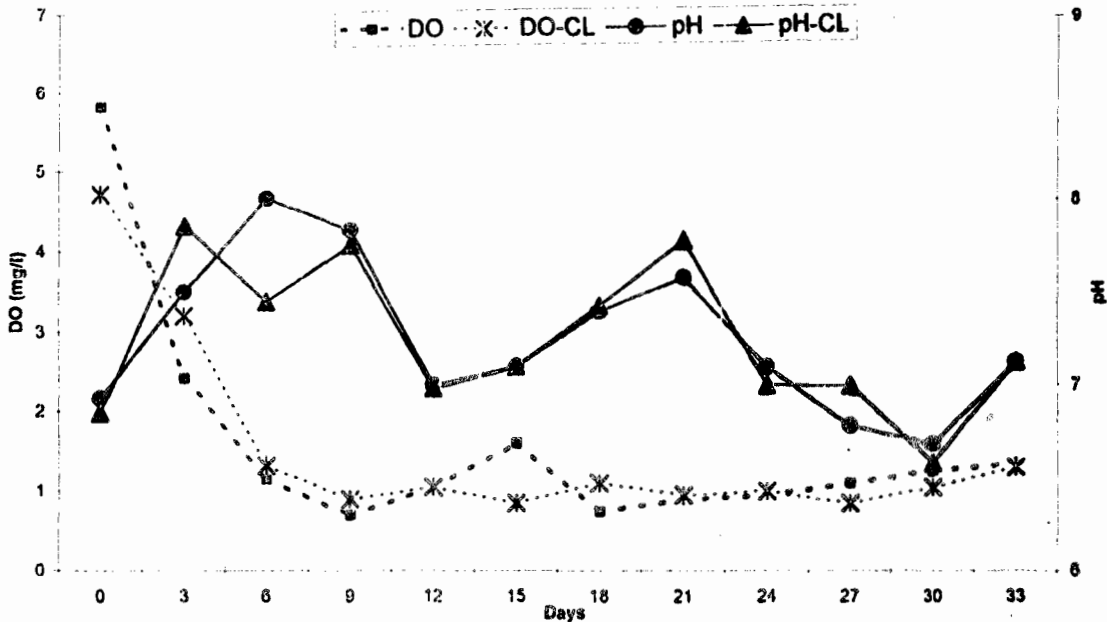


Fig.3 pH and dissolved oxygen concentration changes in treatment and control tanks

consistently to ~25% at day 15. Six days after second application it almost zeroed. A sharp 42% increase by day 27 decreased to less than 10% at termination. Similar inverse relationship was observed between  $\text{NH}_3\text{-N}$  and  $\text{NO}_2\text{-N}$  for the control (Fig. 2) but fluctuated differently.  $\text{NH}_3\text{-N}$  recorded the highest (35% - 73%) proportion except day 24 (34%) and day 27 (38%).  $\text{NO}_2\text{-N}$  followed a similar but inverse trend.  $\text{NO}_3\text{-N}$  fluctuated markedly with the highest of about 45% recorded by day 27. Water quality parameters of pH and  $\text{DO}_2$  changes for the period of experiment is presented as Fig. 3. The pH attained post application maxima of 8.1 and 7.6 in 6 days followed steep decline to 6.8 and 6.5 respectively. The control fluctuated along similar increase-decrease regimes with minimum and maximum of

6.5 and 7.8.  $\text{DO}_2$  fluctuated along similar pattern for the treatments and control with sharp post fertilization decline which remained low except at day 15 for the treatment (1.30mg/l). Both recorded marginal differences between treatment and control means without significant t and F values (Table 1).

The relative plankton abundance for the treatment and control tanks is presented in Figs. 4 and 5. Total of seven taxa comprising four phytoplankton of Desmidiaceae (desmids), Bacillariophyceae (diatoms), Cyanophyceae (blue-green algae) and Chlorophyceae (green algae) and three zooplankton of Protozoa, Cladocera and Rotifera were identified. Phytoplankton was dominated by green algae with maximum relative abundance (~ 70%) after

second fertilizer application, however desmids attained dominant status of 54.5% abundance in the last six days. The blue-green algae were the least represented with a maximum percentage abundance of 6%. Diatoms recorded moderate abundance with range of 10% - 35%. Zooplankton fluctuated greatly with clear dominance by protozoa with maximum of 24% relative abundance. It was followed by rotifers (11%) and cladocerans (6%). The control was alternately dominated by green algae with a range of 3% - 65% and desmids the concluding nine days with a range of 13% - 46% (Fig. 5).

While the blue-green algae was similarly the least represented (~12%). The diatoms recorded reduced relative abundance, a maximum of ~10%. The protozoans also dominated zooplankton attaining maximum abundance of 21% at termination. Significant differences were observed among the plankton group (Fig. 6).

DISCUSSION

Fertilization primarily increases productivity by release of soluble nutrients of which nitrogen in different forms is a known part of these processes

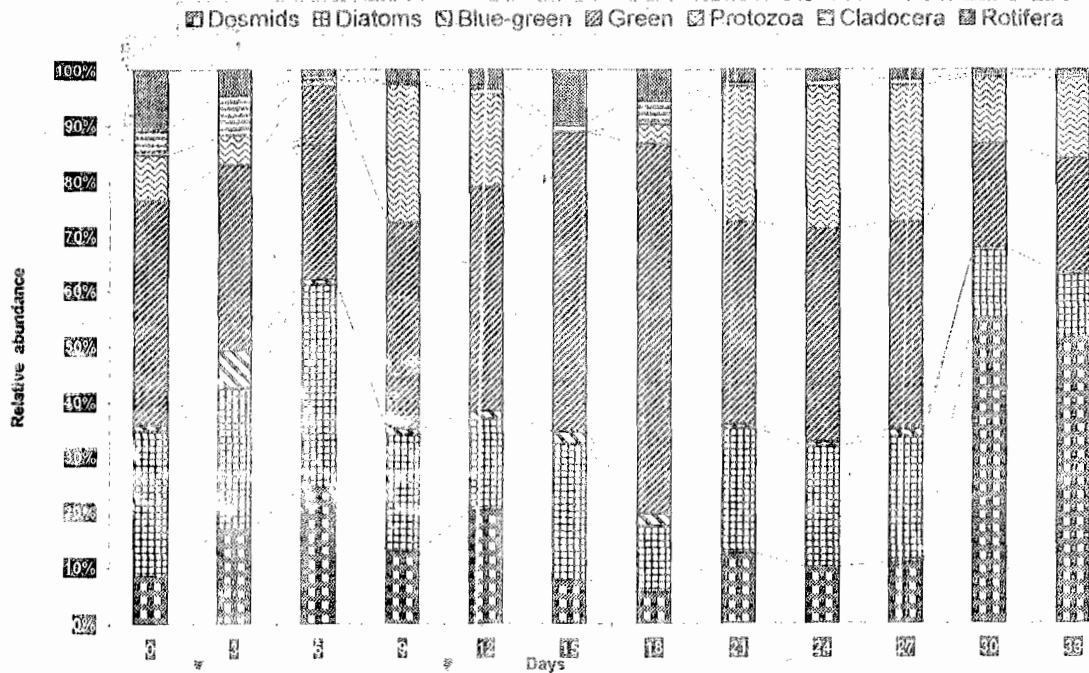


Fig.4 Relative plankton abundance for treatment tanks

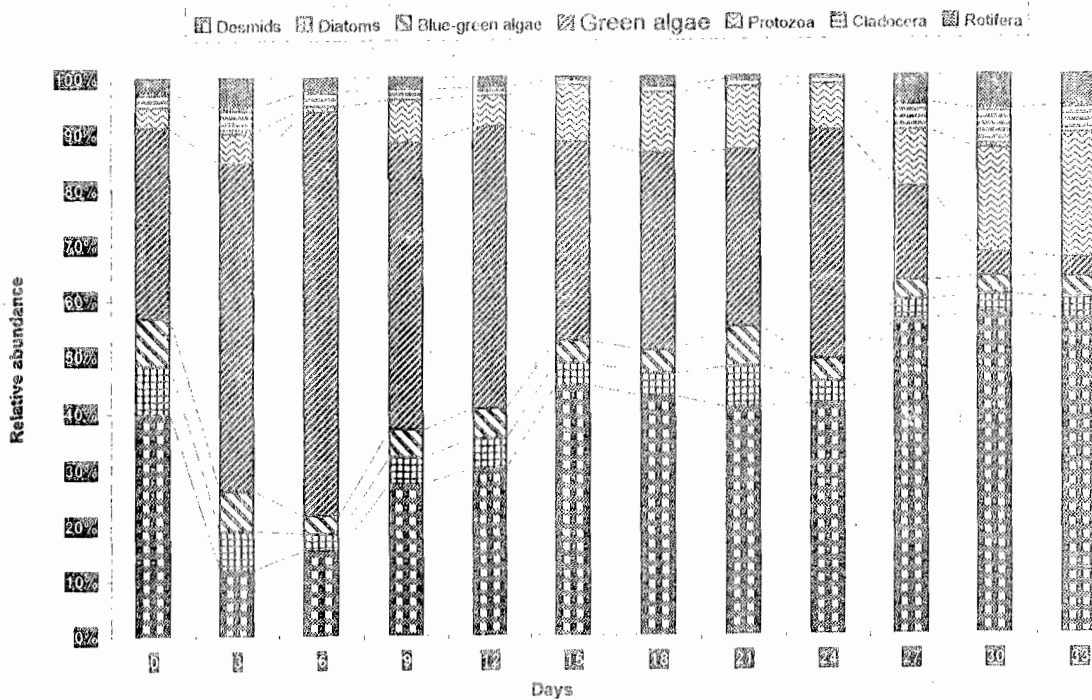


Fig.5 Relative plankton abundance in control tanks

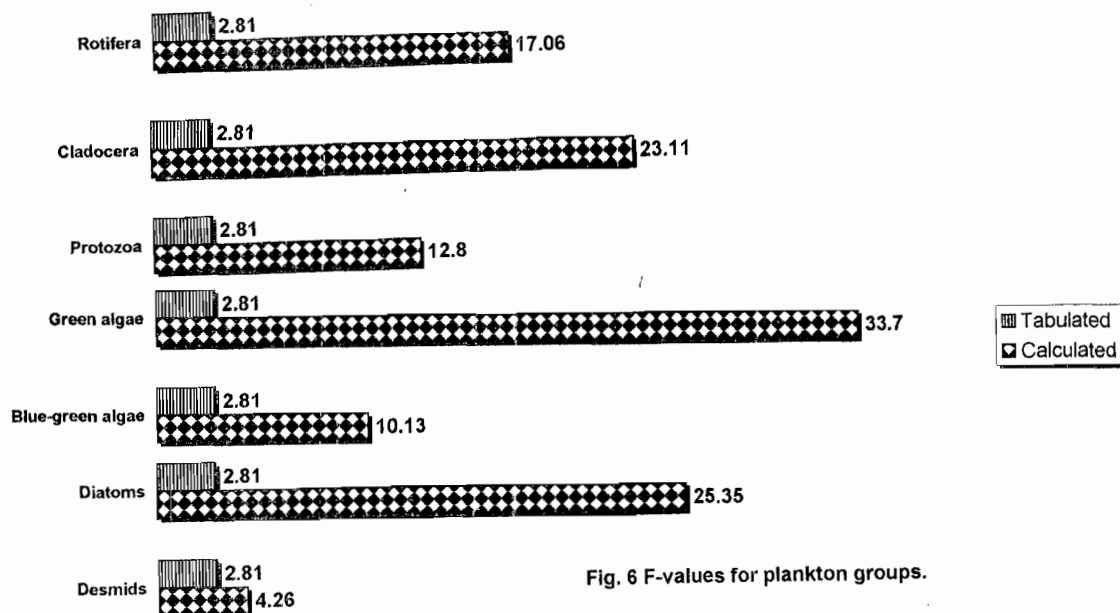


Fig. 6 F-values for plankton groups.

leading to higher productivity. From our results nitrogen forms in treatment tanks recorded increases and declines in concentrations after the fertilization, in agreement with observations of Boyd (1979). The concentration of  $\text{NO}_2\text{-N}$  affects the concentrations of  $\text{NH}_3\text{-N}$  and  $\text{NO}_3\text{-N}$  in the biologically mediated processes, catalyzed via two nitrite intermediate steps in presence of oxygen (Boyd, 1979; Falkowski and Raven, 1997). This transformation was highly favoured by the suitable pH and temperature. Denitrification by heterotrophic bacteria and phytoplankton especially the diatoms with corresponding periods of high density and low DO generated unusual concentrations of  $\text{NO}_2\text{-N}$  and  $\text{NH}_3\text{-N}$  by high rate of biological decomposition in treatment tanks respectively. The observed accumulation of  $\text{NH}_3\text{-N}$  in the control tanks could have similarly, resulted from higher rate of biological decomposition compared with biological recruitment resulting from reduced nutrient availability. This probably accounted for the low DO in the control tanks. Thus differences in statistical significance of nutrient and water quality parameters.

The variations in plankton density indicated clearly the influence of external nutrient inputs (Jeje, 1986; Brummet, 2000; Jeje and Sowunmi, 2000) and Rabalais (2002) gave excellent examples of studies that reported significant positive relationships between nutrient loading and algal biomass. This accounted for the significant differences across treatments and controls. The relative abundance of each taxa reflected the ability to utilize  $\text{N}_2$ , in addition to other necessary nutrients; while the zooplankton reflected the presence and utilization of food organisms. The dominance by non-nitrogen fixing algae (i.e. desmids, diatoms and green algae) indicated that the tanks were not nitrogen restricted, in contrast to the control which had no

additional  $\text{N}_2$  inputs hence reduced N:P thus favoured dominance by blue-green algae (Figs. 4&5), similar to observations of Smith (1983) and Paerl and Tucker (1995). However, the relative phytoplankton abundance in the control especially for desmids and green algae appeared to be near maximum suggestive of nutrient patchiness from zooplankton excretion as has been reported by Sterner (1986) and Kilham and Hecky (1988). Rabalais (2002) in her review noted this feature of uncommon singular nutrient limitation in freshwater system compared with marine systems. In both the treatment and control tanks all the zooplankton taxa fluctuated along similar pattern i.e. Protozoa, Cladocera and Rotifera with treatment tanks recording higher abundances. According to Infante and Edmonton (1985) and Rothhaupt (1995) structure and dynamics of zooplankton are highly dependent on the availability and quality of edible phytoplankton species. The available phytoplankton taxa with exclusion of blue green algae were among those generally considered to be of good quality to zooplankton (Infante and Edmonton, 1985; Paerl and Tucker, 1995; Rothhaupt, 1995). However, in this study an additional factor will have to be considered: algal nutrient limitation. Rothhaupt (1995), established a reduction in zooplankton growth rate in nutrient limited environment and concluded that nutrient limitation greatly altered food quality hence mineral nutrient in the zooplankton.

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