



The Response of Phytoplankton Growth to Nitrogen and Phosphorus; Focus on Seston Biomass and its Implication for Fish Pond Fertilization

Marshet Adugna Mitiku

Ethiopian Institute of Agricultural Research (EIAR),
National Fishery and Aquatic Life Research Centre (NFALRC)
Email: marshetadu@gmail.com

Abstract

Phytoplankton productivity can be affected by nutrient availability such as phosphorus and nitrogen. The study was conducted to test the effects of nitrogen and phosphorus alone and in combination on seston biomass change in experimental tanks. Four treatments were used (control, phosphorus, nitrogen and phosphorus plus nitrogen) to see nutrient effect on seston biomass. Dissolved oxygen and turbidity were high in nitrogen and phosphorus (NP) treated group. The addition of nitrogen and phosphorus together has a significant effect on the of seston biomass than nitrogen and phosphorus treated tank ($p < 0.05$). Phosphorus treated tank (P) showed better seston ash free dry weight, primary productivity and chlorophyll *a*.

Keywords: Seston, phytoplankton, primary productivity, phosphorus, nitrogen,

Introduction

The most important component of aquatic food webs are phytoplankton and they transfer both nutrients and energy to higher trophic levels (Reynolds, 2006). Phytoplankton cells require elements in order to grow and reproduce. Application of phosphorus and nitrogen increases in phytoplankton productivity and in an increase of chlorophyll *a* and its biomass. Many factors play an important role in determining phytoplankton productivity in aquatic systems such as light, temperature and nutrients (Yusoff, F. M. *et al.*, 1989). The seston are suspended particulate matter in water including living, detritus and inorganic particles largely vary in size, abundance and nutritional value. They are important source of energy at lower trophic levels and secondary production of zooplankton and fish (Schindler, D. W., 2006).

Phosphorus and nitrogen has an important role in primary productivity of phytoplankton. Their effect on the primary productivity at natural water bodies was studied by many researchers (Schindler D.W., 1977). However, the effect of these nutrients s not well studied on the effect of seston biomass. Whether the growth of the seston community be increased or decreased significantly by the addition or removal of nitrogen or phosphorus needs to be addressed. This study have focused on the type of nutrient which has the greatest effect on the seston biomass in terms of variables like primary production, chlorophyll *a* concentration as surrogate for biomass, net and gross primary productivity and their response to nutrient enrichment. In addition, physical properties of the water such as turbidity as it affects light penetration and dissolved oxygen were measured to relate with

photosynthetic activity. The objective of this study was to test the effects of nitrogen and phosphorus on seston biomass in experimental tanks.

Materials and Methods

Study site

The study was conducted at National Fishery and Aquatic Life Research Centre in Sebeta, Ethiopia located. Water sample was taken from fish research ponds NFALRC-ponds and experimental mesocosms were set.

Study design

The study was carried out in September, 2019 at EIAR-NFALRC. A factorial experiment was designed to test the response of nitrogen and phosphorus on seston biomass. Sixteen plastic containers (65×45×40cm) were placed on the ground with 4 treatment combinations (Table 1). Each container was filled with 86 litres of tap and pond water. For three consecutive weeks, 50 ml solutions of phosphorus and nitrogen were added once per week with 16:1 ratio.

Physico-chemical parameters

Water quality were parameters such as pH, temperature, conductivity and dissolved oxygen were measured in situ using their respective meters. Water sample was taken to measure turbidity in terms of light absorbance as a surrogate for light absorption by a spectrophotometer.

Seston biomass measurement

For analysis of chlorophyll *a*, a 1000 ml volume of water sample was filtered on to a Schleicher & Schuell GF 6 glass fibre filter of pore size 0.2 µm by the vacuum filtration set-up. Chlorophyll *a* was extracted from filter in 25 mL 85% ethanol at 75 °C for 5 minutes. The extract was then analysed by spectrophotometer at 750 nm for chlorophyll *a*.

Seston ash free dry weight was measured by filtering 1000 ml of sample water for each sample and dried for 24 hours in oven at 105 ° C. The dry weight of the sample was determine. The dried sample was then combusted at 520°C to reduce it to mineral ash for calculation of the ash free dry weight seston biomass

and is determined using the standard methods of laboratory manual (Gettel. *et.al.*, 2016).

Primary productivity of seston was measured by measuring the rate of oxygen production in the light and the rate of oxygen consumption in the dark. For each treatment, two gas-tight Winkler bottles were filled with water sample and initial oxygen concentration in each bottle was measured. One bottle in the light using a high intensity lamp as light source and one bottle in the dark wrapped with aluminium was placed. After 2 hours, final oxygen concentration in each bottle was measured. A gross primary production rate was calculated by summing the oxygen produced during the light with the oxygen consumed during the dark where as net primary productivity is measured as the oxygen produced in the light bottle (Gettel. *et.al.*, 2016).

Statistical analysis

Normality of variances as tested by applying the Shapiro Wilke test. Measures analyses of variance (ANOVA) was used to quantify the individual and combined effects significance of nutrient availability on dissolved oxygen, turbidity, ash free dry weight, chlorophyll *a* concentrations and primary productivity. Data was organised in Ms excel and all statistical analyses were analysed in the statistical program R version 3.3.0.

Results

The concentration of dissolved oxygen in the experimental tanks was found to be higher in a treatment having both nitrogen and phosphorus (Figure 1a). Differences in oxygen concentration between the treatments was significantly different ($p=0.007$). This difference is mainly attributed to the treatment with nitrogen and phosphorus ($21.5\pm 2.2\text{mg/L}$) and the one with nitrogen only ($13.2\pm 1.4\text{mg/L}$).

Turbidity was measured in terms of light absorbance and highest in control tank ($97.3\pm 1.0\%$) which is cleaner and is lowest ($74.3\pm 2.6\%$) in a tank treated with both nitrogen and phosphorus (Figure b). This was significantly different between treatments ($p=0.0004$) and the percentage was highly significant between control (C) and nitrogen and phosphorus treatment group (NP).

Seston biomass in terms of ash free dry weight was found to be higher in the treatment group with both nitrogen and phosphorus (27.6±6.8 mg/L). It was lowest in the control group (2.4±1.1 mg/L). The difference between the treatments was significantly different ($p= 0.003$).

Seston chlorophyll *a* was found to be far highest in tanks in which both phosphorus and nitrogen were added, with a mean value of 222.37±78.2mg/L. The treatment which received nitrogen only and the control group had the lowest mean chlorophyll *a* of 3.43±1.4mg/L and 4.36±2.4 mg/l respectively. Differences between treatments were significant at $P< 0.05$ (Figure 1. Mean net primary productivity as estimated by the light-dark bottle oxygen method was highest for nitrogen phosphorus combination (NP) treatment (2.16± 0.2(mg/L).The treatments were significantly different at $p=0.003$ (Figure 1d).

As primary productivity, ponds treated with both phosphorus and nitrogen produced the highest mean gross primary productivity (32.62±5.1mg/L). It is followed by phosphorus treated, nitrogen treated and control with 22.61±2.7 (mg/L), 19.20±2.6 (mg/L), and 10.66±4.8(mg/L)respectively (Figure 1f). The difference between the treatments was significant ($p<0.015$) (Figure 1f).

Seston ash free dry weight followed the similar pattern as chlorophyll *a* with the highest values occurring in the same treatments. Net primary productivity and gross primary productivity followed the same patterns in the same treatments from control, nitrogen, phosphorus and nitrogen phosphorus combination occurring in increasing order (Figure1, e and f).

Table 1. A microcosm experiment in the compound of NFALRC-Sebeta experimental design

Treatment	Number of containers	Nitrogen(N)	Phosphorus(P)
1	4	+	+
2	4	-	-
3	4	+	-
4	4	-	+

Table 2. One way ANOVA test results of selected variables showing f and p values

	Df	Sum Sq	Mean Sq	F-value	Pr(>F)
DO	3	161.6	53.85	6.41	0.008
Turb	3	1194.5	398.2	12.74	0.0005
AFDW	3	1436	478.6	7.94	0.003
Chl- <i>a</i>	3	1413	471	7.69	0.003
NPP	3	20	6.66	8.675e+31	<2e-16
GPP	3	989.72	329.91	5.2089	0.016

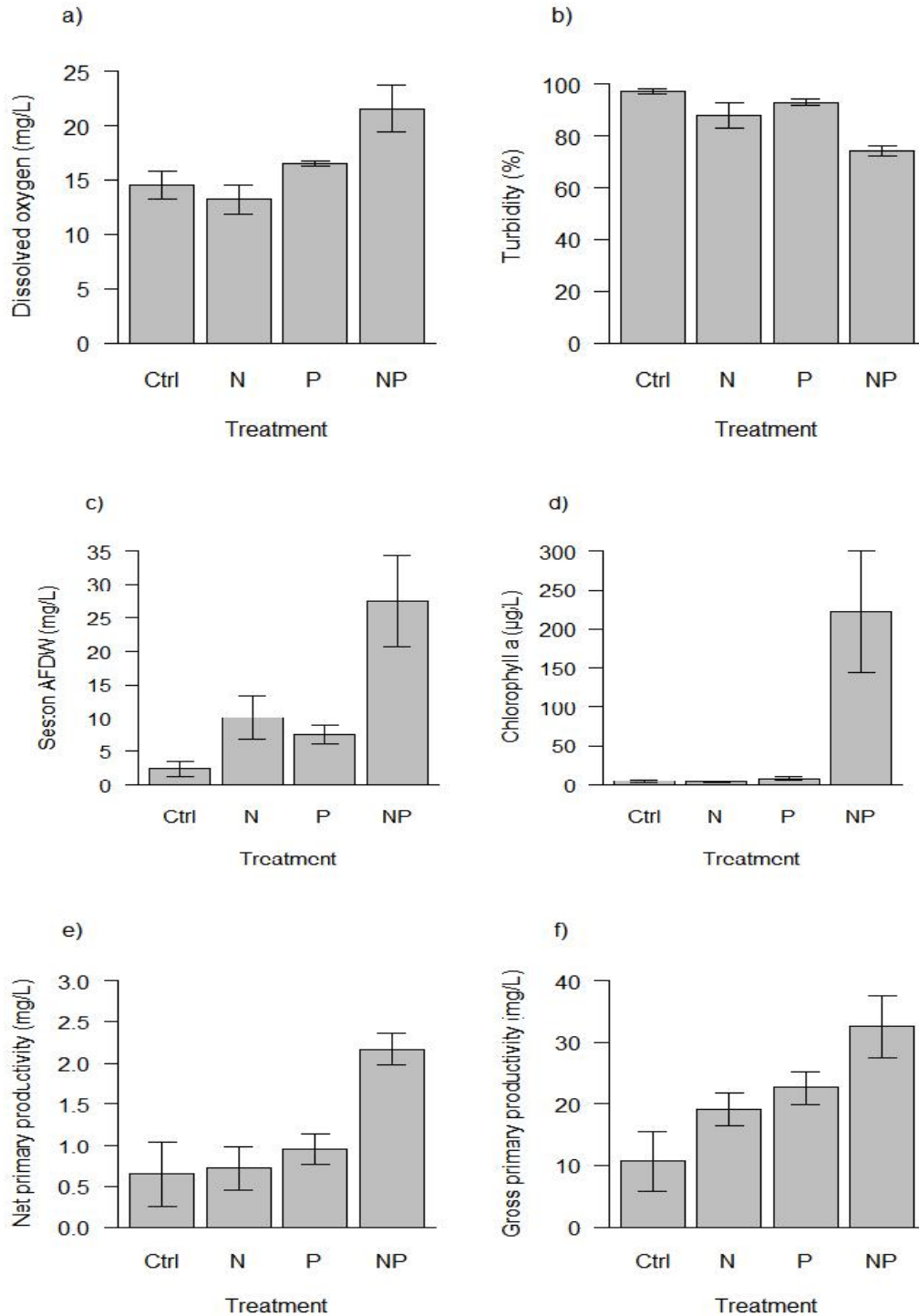


Figure 1. Effects of phosphorus and nitrogen on (a) Dissolved oxygen, (b) Turbidity, (c) Seston ash free dry weight(AFDW), (d) Chlorophyll a concentration, (e) Net primary productivity, and (f) Gross primary productivity. Means and standard errors are depicted for each treatment (n=4).

Discussion

Phytoplankton undergo photosynthesis which adds oxygen to the water system. The highest dissolved oxygen concentration in the treatment group having nitrogen and phosphorus might probably be due to high seston biomass associated with the presence of the two essential nutrients which helps for the growth of seston as shown in figure 1c.

The light absorbance was found to be lowest in the nitrogen and phosphorus treated group which shows high turbidity. Seston ash free dry weight, chlorophyll *a* and primary productivity were highest in these treatment group, (Figure 1 c, d, and e) which indicate that seston biomass contribute increased turbidity. Seston biomass was lowest in control group, which also had high light absorbance, this indicated that lower biomass causes high light penetration. Despite presence of light in the control which helps in phytoplankton carbon (C)-fixation rates, leading to increased phytoplankton C and hence seston growth, (Elser and Urabe, 1999), absence of nutrients in the control, caused lower seston biomass that was observed.

Addition of phosphorus and nitrogen together to water increased seston ash free dry weight, chlorophyll *a* and both net and gross primary productivity. This shows that both nitrogen and phosphorus are likely important to make unproductive water system more productive than adding individual nutrient addition. A study conducted by Diana *et al.*, (199) showed the same result in a study conducted to see the effect determinant chlorophyll *a* that phosphorus was a key determinant of sestonic chlorophyll *a*.

Gross and net primary production showed trends similar to chlorophyll *a*, with significant differences between tanks for each treatment. The same result was found by Elser *et al.*, (2007) that primary production showed similar trend like chlorophyll *a*. However, addition of phosphorus only increased chlorophyll *a* and primary productivity, which was relatively higher than when nitrogen alone is added. Similar result also showed from bioassays on phosphorus limited of low productive water systems (Payne, A. G., 1975).

Conclusions and Recommendations

From this study, dissolved oxygen and turbidity of the water treated with both nitrogen and phosphorus was highest than other treatment groups. Addition of

nitrogen and phosphorus in together has a significant effect on the seston biomass as depicted by chlorophyll *a*, ash free dry weight and primary productivity. Phosphorus treated tank has better growth than nitrogen as shown in primary productivity and chlorophyll *a*. The limiting nutrient in this experiment was phosphorus. But the experiment needs extended time and many replicates to see the effect of these nutrients on phytoplankton growth. Therefore, both phosphorus and nitrogen should be applied if production of seston is needed. Nutrient release to water system should be monitored to control eutrophication which could affect fish growth and health. Further study should be conducted with many replicas and in the natural water canals for better understanding of the effect of these two nutrients on seston biomass.

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