

## Influence of inorganic fertilizer application and supplementary feeds on periphyton biomass, quality and species composition

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### Abstract

A study was conducted to assess the effects of fertilization alone (T<sub>1</sub>), supplementary feeding alone (T<sub>2</sub>) and combination of fertilization plus supplementary feeding (T<sub>3</sub>) on periphyton species composition, quantity and quality. Sex-reversed *Oreochromis niloticus* fingerlings were stocked in earthen ponds one week after initial pond fertilization. Urea and Diammonium phosphate (DAP) fertilizers were applied in pond water at weekly intervals at a rate of 3 and 2 g/m<sup>2</sup>, respectively. Fish were fed feed containing 25.1% crude protein at 5% (T<sub>2</sub>) and 2.5% (T<sub>3</sub>) of fish body weight daily. The results show that the ponds under combination of fertilization plus supplementary feeding (T<sub>3</sub>) had higher periphyton biomass (47.35 ± 7.64 g DM/m<sup>2</sup>), crude protein content (11.40 ± 0.16%) and organic matter content (OM) (25.47 ± 0.28%). The periphyton from ponds under fertilization alone (T<sub>1</sub>), had the highest ether extract content (1.84 ± 0.07%) and ponds treated with supplementary feeding alone (T<sub>2</sub>) had higher phosphorous content (0.48 ± 0.0 mg/L). The body of fish cultured in ponds under T<sub>3</sub> had higher CP (69.14 ± 0.33%) and OM (96.65 ± 0.16%) contents while those reared under T<sub>1</sub> had higher ether extract content (18.33 ± 0.19%) and ash content (4.78 ± 0.1%), suggesting a positive relationship between algal quality and fish muscles. In addition, fish growth rate increased as the periphyton quantity (biomass and OM) and quality (CP and phosphorous) increased. The study also revealed five classes of phytoplankton (Bacillariophyceae, Chlorophyceae, Cyanophyceae, Euglenophyceae and Zygnematophyceae) and three classes of zooplankton (Eurotatoria, Heterotrichea and Oligohymenophorea). In general, both phytoplankton (algae) and zooplankton were more abundant in ponds under T<sub>3</sub> than in ponds under the other treatments. It is concluded that the combination of fertilization plus supplementary feeding (T<sub>3</sub>) produces higher periphyton quantity, quality and species composition and thus promotes higher fish growth rate compared to feeding alone and fertilization alone treatments.

**Keywords:** algae, growth rate, Nile tilapia, proximate chemical composition, zooplankton,

### 1. Introduction

Fish growth performance is a function of the genetic make up and exogenous factors, particularly nutrition and management [1]. Fish grown in ponds can be fed supplementary feeds or natural food produced through fertilization [2,3]. Fertilizer application in pond water supplies essential nutrients, especially nitrogen (N) and phosphorous (P) which are the most limiting nutrients for algae (phytoplankton) growth in freshwater ecosystem [3]. Moreover, the addition of supplementary feeds in pond water increases nitrogen and phosphorous contents from the decomposition of uneaten feeds which, in turn, results into higher quantity of plankton community in the pond ecosystem [4]. The concentration of plankton in the water column determines the amount of periphyton (phytoplankton and zooplankton attached into the substrate). Phytoplankton and zooplankton stabilize the aquatic ecosystem and minimize the fluctuations of water quality through photosynthesis, respiration and assimilation of ammonia [5]. In addition, periphyton supply some important enzymes that improve the utilization of the supplemented feed, thus increase feed utilization efficiency [6].

The quantity and quality of periphyton improves fish growth rate and biochemical composition of the fish [7]. The periphyton quantity and quality can be manipulated through fertilization and management of water quality [8]. The quality of algae (phytoplankton) is influenced by the concentration

of nitrogen (N) and phosphorus (P) in pond water, rather than energy (carbon). The presence of carbon (C) in the cell wall of algae lowers digestibility of periphyton [9,10]. The major component for algal growth is nitrogen due to its function in the structural composition of cells and functional proteins such as enzymes in algal cells [11].

Lipids from natural food are the imperative nutrients for fish as they contain essential fatty acids (EFAs) and energy which are required for fish growth and healthy tissue maintenance. In changes in nitrogen affect the availability of essential fatty acids which includes docosahexaenoic acid (DHA) and eicosapentaenoic acid (EPA). These essential fatty acids must be provided in the diet because they cannot be synthesized by the fish body [12, 13, 14]. Also, zooplankton as a constituent of natural food has been reported to contain high crude protein and lipid contents which are important for fish growth [15].

Biomass and community composition of the periphyton differs depending on nutrients availability, water quality and grazing pressure caused by other aquatic organisms (top down effects) [2, 3]. Higher plankton quality and biomass in ponds may lead to improved fish growth performance [2]. The presence of high quality natural food in a fertilized pond production system is vital for fish growth, because the availability of high quality natural food in pond water minimizes the dependence on supplementary feed, hence, reduces the production costs and increases profit. This is desirable, especially for smallholder farmers who have low

capital [16].

A study done in Egypt to assess the effect of supplementation of fertilized ponds cultured with tilapia showed that the combination of supplementary feeding plus fertilizer application results into more plankton abundance and species composition than fertilization alone or feeding alone [2,16]. In Tanzania, the information on the effects of inorganic fertilizer application and supplementary feeds on periphyton quantity, quality and species composition is lacking. This study, therefore, was conducted to assess the effects of inorganic fertilizer application alone, feeding alone and their combination on periphyton quality, quantity and their influence on fish muscles biochemical composition.

## 2. Materials and Methods

### 2.1 Description of the study location

The experiment was conducted in earthen ponds for 180 days at Kilosa district, Tanzania. Kilosa district lies between latitude 5° 55'S and 8° 53'S and longitude 36° 30'E and 37° 30'E. The area of experiment is subject to seasonal flooding and characterized by poorly drained soils with black cracking clays during the dry period. The district receives rainfall between October and May with the mean annual rainfall ranges from 800 to 1400 mm [17].

### 2.2 Management of ponds and fish

The experiment was conducted in nine earthen ponds, each with an average area of 177 m<sup>2</sup>. The treatments were fertilization alone (T<sub>1</sub>), supplementary feeding alone (T<sub>2</sub>) and the combination of fertilization plus supplementary feeding (T<sub>3</sub>), each treatment was replicated three times. Urea and Diammonium phosphate (DAP) fertilizers were applied at weekly intervals in ponds under treatments T<sub>1</sub> and T<sub>3</sub> at the rate of 3 g and 2 g per m<sup>2</sup>, for Urea and DAP, respectively. Sex-reversed Nile tilapia fingerlings with weight ranging from 0.7 to 1.1 g were collected from Ruvu fish farm and stocked seven days after initial pond fertilization at a stocking density of 3 fingerlings /m<sup>2</sup>. Feeding in treatments T<sub>2</sub> and T<sub>3</sub> was done twice per day using a diet containing 25.1% crude protein (CP). Fish were fed at the feeding level of 10% and 5% of the fish body weight (FBW) for T<sub>2</sub> and T<sub>3</sub>, respectively, for the first two months. Then the amount of feed was reduced to 5% and 2.5% for T<sub>2</sub> and T<sub>3</sub>, respectively, for the last four months of the experimental period.

### 2.3 Fish sampling and growth rate determination

Fish were sampled biweekly using a net with 1 mm mesh size. A random sample of 30 fish was taken from each pond and each fish was individually measured for body weight (g) and length (cm), using a portable digital weighing balance and board fixed with a ruler respectively. After taking measurements the fish were released back into their respective ponds. The growth rate (GR) was calculated using the following formula [18, 19]:

$$GR = \frac{\text{Final weight (g)} - \text{initial weight (g)}}{\text{Experimental period (days)}}$$

Where: GR is growth rate

### 2.4 Determination of chemical composition of fish carcass

At the end of the experiment, three fish from each pond were collected, put in the ice box and transported to a

laboratory and stored in the deep freezer at -18 °C. Then, the fish were thawed at room temperature, cleaned, eviscerated, filleted and deboned [20, 21]. The fish were dried at 60 °C for 24 hours. The dried fish samples were homogenised by grinding in the motor to an average sieve size of 2 mm, and then put in glass bottles for dry matter, ash, crude protein and ether extract determination [22]. Crude protein (CP) was determined by Kjeldahl method [22, 23, 7, 24]. Ether extract was determined by the Soxhlet extraction method [22, 24].

### 2.5 Periphyton quantity, quality and community composition analysis

Eight nets, each with 20 µm-mesh size and an area of 1250 cm<sup>2</sup> were placed (fully submerged in water) in each pond for periphyton to attach. After every two months, the nets were taken out from the water and scrubbed to collect algae and zooplankton. Four ml of the periphyton solution were taken and preserved in 4% concentration of formalin solution for species composition identification. In the laboratory 10 µL of the periphyton sample, in duplicate, was taken by using a pipette and placed in the neubor chamber slide for enumeration by using microscope at 10x magnification. For each treatment the number of zooplankton and phytoplankton were determined and expressed as a mean number of taxa [25]. The rest of the scrubbed periphyton samples were allowed to settle and the water was decanted to concentrate the periphyton. Samples were put in 500 ml bottles, transported in an ice cool box to a laboratory and stored in deep freezer at -18°C for laboratory analysis. In the laboratory periphyton samples were centrifuged at 3000 rpm for 10 minutes, then dried in an oven at 60°C for 24 hours for biomass and proximate chemical compositions analysis [26, 22]. Duplicate sub-samples, each weighing approximately two grams, were taken and burned for 3 hours at 550 °C and digested in 6 Normality of HCL for phosphorous determination using Spectrophotometer analysis. The absorbance of the samples and standard solution were read in the Spectrophotometer at the wavelength of 420 nm [27].

### 2.6 Statistical analysis

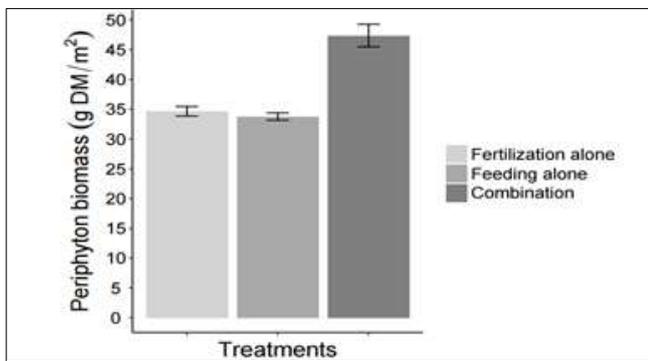
Data were analysed by using R Studio software version 3.5.0 (2018) [28, 29]. Before the analysis of variance, the data were checked for normality and transformed whenever necessary to increase error homoscedasticity. One-way ANOVA was used at 5% significance level to assess the effect of treatment on periphyton quantity (biomass, DM, OM), quality (CP, EE, and Phosphorous) and fish body chemical composition. Post Hoc analysis was done by using Duncan's new multiple range test to determine the significance of the differences between treatment means when the treatment had significant effect. In addition, correlation was used to determine the relationships between periphyton proximate chemical composition and fish growth and muscles proximate chemical composition values. Multiple regression analyses were used to assess the relationship between periphyton proximate and fish growth performance. The model for multiple regression analyses was as follows:

Where: = Response (fish growth rate) = The intercept of the regression line on Y axis k<sup>th</sup> partial regression coefficient (slope), ... = k<sup>th</sup> predictor variable [periphyton proximate chemical composition (i.e. Biomass, CP, EE and phosphorous)] = error term

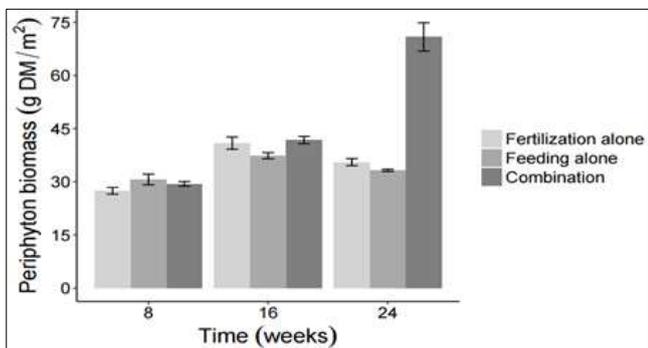
### 3. Results

#### 3.1 Periphyton Biomass

Results on biomass of periphyton found in the ponds under different treatments are shown in Fig. 1 and Fig. 2. Periphyton biomass differed significantly ( $p < 0.05$ ) among the treatments. The ponds subjected to combination of fertilization plus supplementary feeding ( $T_3$ ) had higher mean periphyton biomass ( $47.35 \pm 7.64$  g DM/m<sup>2</sup>) than those under feeding alone and fertilization alone (Fig. 1). However, the means of periphyton biomass between ponds under fertilization alone ( $34.64 \pm 3.21$  g DM/m<sup>2</sup>) and feeding alone ( $33.76 \pm 2.43$  g DM/m<sup>2</sup>) did not differ ( $p > 0.05$ ). The mean periphyton biomass from the ponds under the combination of fertilization plus supplementary feeding treatment increased as the experimental time increased while for the other treatments periphyton biomasses decreased toward the end of the experiment (Fig. 2).



**Fig 1:** Comparison of periphyton biomass (Mean ± se) in ponds under fertilization alone, feeding alone and combination of fertilization plus supplementary feeding”

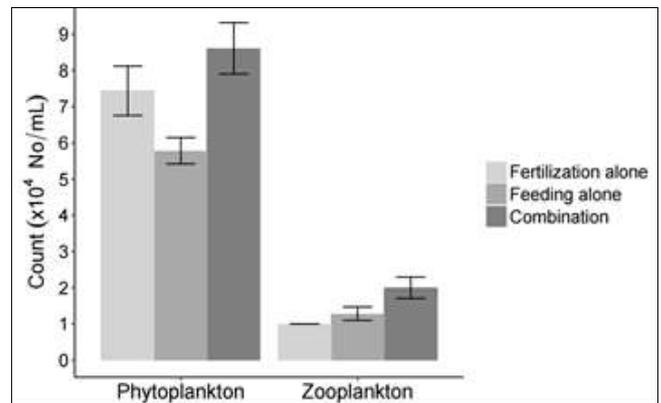


**Fig 2:** Variation in periphyton biomass (Mean ± se) over time in ponds subjected to fertilization alone, feeding alone and the combination of fertilization plus supplementary feeding

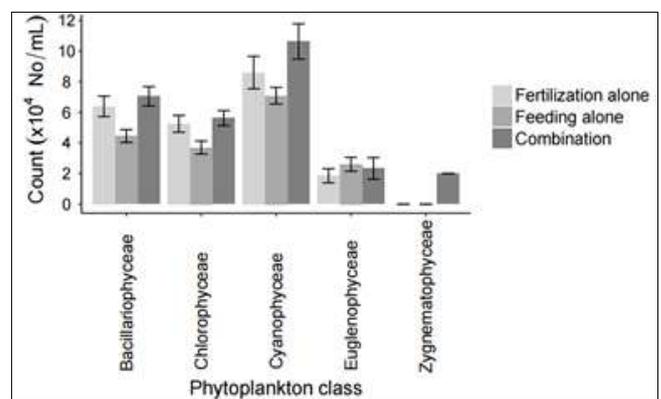
#### 3.2 Periphyton species community composition and abundance

Results for community composition and abundance of periphyton species from ponds under fertilization alone, feeding alone and the combination of fertilization plus supplementary feeding are shown in Fig. 3, Fig. 4 and Fig. 5. The results show that there was significantly higher phytoplankton abundance than zooplankton abundance (Fig. 3). The ponds under the combination of fertilization plus supplementary feeding had higher number of both phytoplankton and zooplankton than the ponds under the other treatments. The ponds under feeding alone treatment showed the lowest phytoplankton abundance while those under fertilization alone treatment had the lowest zooplankton

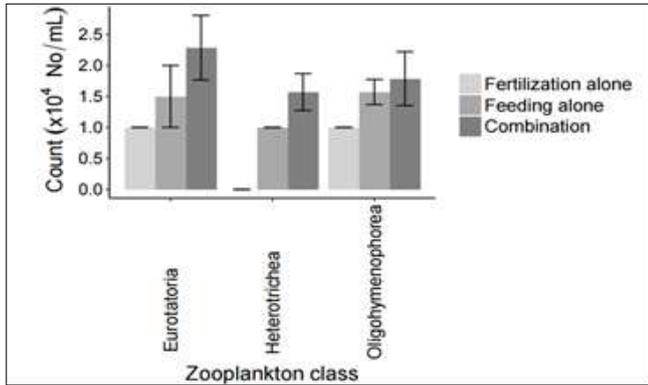
Abundance (Fig. 3). The phytoplankton classes found in the experimental ponds, with the number of recorded genera in bracket, were Bacillariophyceae (2), Chlorophyceae (3), Cyanophyceae (9), Euglenophyceae (4) and Zygnematophyceae (1) (Fig. 4). The dominant genera in each class were as follows: *Frustulia* and *Nitzschia spp.* for Bacillariophyceae, *Closterium sp.* for Chlorophyceae, *Anabaene*, *Microcystis* and *Planktothrix spp.* for Cyanophyceae, *Phucus sp.* for Euglenophyceae and *Staurastrum sp.* for Zygnematophyceae. Among the phytoplankton classes in the present study, Cyanophyceae had the highest abundance in all treatments while Zygnematophyceae class was only observed in ponds under the combination of fertilization plus supplementary feeding. The zooplankton classes observed in the present study, with the number of recorded genera in bracket, were Eurotatoria (2), Heterotrichea (2) and Oligohymenophorea (4) (Fig.5). The dominant genera observed under each class were *Lecane sp.* for Eurotatoria, *Spirostom sp.* for Heterotrichea, *Carchesium* and *Lembadion spp.* for Oligohymenophorea. Heterotrichea was observed in ponds under the treatments that included either feeding alone or combination of fertilization plus supplementary feeding. Heterotrichea exhibited significantly higher abundance in ponds under the treatment for combination ( $T_3$ ) than in ponds under feeding alone ( $T_1$ ) (Fig.5). In general, the ponds under the combination of fertilization plus feeding showed the highest abundance of zooplankton, followed by those under feeding alone ( $T_2$ ) and fertilization alone ( $T_1$ ) (Fig. 5).



**Fig 3:** Phytoplankton and zooplankton abundance in ponds under fertilization alone, feeding alone and combination of fertilization plus supplementary feeding



**Fig 4:** Phytoplankton abundance in ponds under fertilization alone, feeding alone and combination of fertilization plus supplementary feeding



**Fig 5:** Zooplankton abundance in ponds under fertilization alone, feeding alone and combination of fertilization plus supplementary feeding

**3.3 Periphyton proximate chemical composition**

Table 1 summarises the mean values for proximate chemical composition of periphyton collected from ponds under fertilization alone, feeding alone and combination of fertilization plus feeding. Periphyton from the ponds subjected to the combination of fertilization plus feeding had significantly ( $p < 0.05$ ) higher CP ( $11.40 \pm 0.16\%$ ) and OM ( $25.47 \pm 0.28\%$ ) contents while periphyton from the ponds under fertilization alone had the least CP content ( $8.97 \pm 0.22\%$ ). The difference in OM content of periphyton in the ponds under fertilization alone ( $23.23 \pm 0.33\%$ ) and those from ponds under feeding alone ( $24.78 \pm 0.41\%$ ) was insignificant ( $p > 0.05$ ). The periphyton ether extract (EE) contents differed significantly ( $p < 0.05$ ) among the treatments (Table 1). The highest periphyton EE value ( $1.84 \pm 0.07\%$ ) was observed in periphyton from ponds under fertilization alone ( $T_1$ ) while the lowest value ( $0.97 \pm 0.05\%$ ) was observed in periphyton from ponds under the combination treatment ( $T_3$ ). The highest phosphorous (P) content was observed in the periphyton from ponds under feeding alone and the lowest was found in periphyton from the ponds under fertilization alone ( $T_1$ )

**Table 1:** Proximate chemical composition (Means  $\pm$  se) of periphyton from ponds under fertilization alone, feeding alone and combination of fertilization plus feeding

Proximate composition	Treatments		
	Fertilization alone ( $T_1$ )	Feeding alone ( $T_2$ )	Combination ( $T_3$ )
DM (%)	$96.27 \pm 0.06^a$	$96.37 \pm 0.08^a$	$95.38 \pm 0.10^b$
CP (%)	$8.97 \pm 0.22^c$	$10.68 \pm 0.25^b$	$11.40 \pm 0.16^a$
OM (%)	$23.23 \pm 0.33^b$	$24.78 \pm 0.41^b$	$25.47 \pm 0.28^a$
EE (%)	$1.84 \pm 0.07^a$	$1.43 \pm 0.08^b$	$0.97 \pm 0.05^c$
P (mg/L)	$0.35 \pm 0.01^c$	$0.48 \pm 0.0^a$	$0.41 \pm 0.0^b$

\* abc= Means with the same superscript letter in the same row are not significantly different ( $p > 0.05$ ).

**3.4 Relationship between periphyton quantity, quality and fish growth rate**

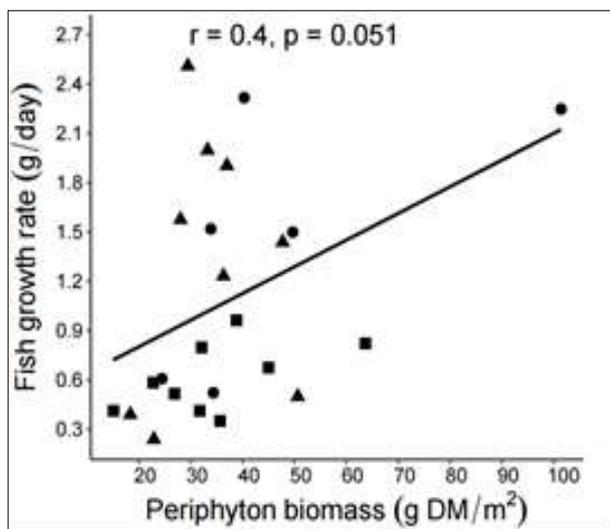
Table 2 present the regression of fish growth rate on periphyton biochemical composition. The results show that the growth rate of fish reared in the ponds under fertilization alone, feeding alone and the combination of fertilization plus supplementary feeding was influenced by periphyton biomass and biochemical composition.

**Table 2:** Regression of fish growth rate on periphyton biomass and proximate chemical composition

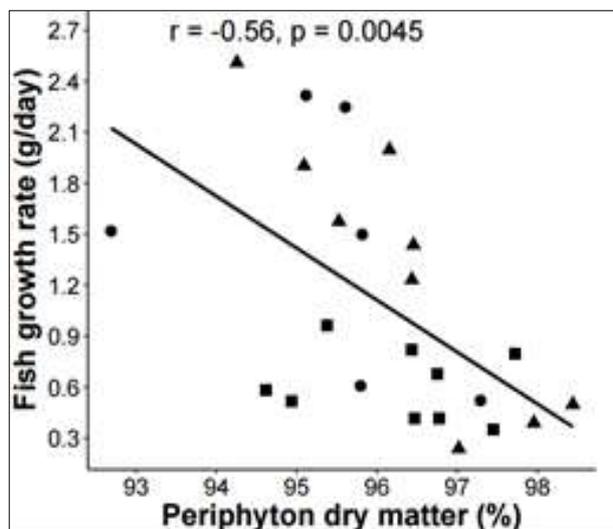
Periphyton variable	Partial coefficient of Regression (b)	se	p-value
Biomass	0.019882	0.003899	0.001
CP	0.110326	0.019560	0.001
EE	-0.025346	0.062457	0.687
Phosphorous	1.518757	0.711005	0.038

Note: CP=Crude protein, EE=Ether extract, se = standard error.

Fish growth rate was positive and significantly ( $p < 0.05$ ) correlated with periphyton biomass, organic matter, crude protein and phosphorous, (Fig. 6 (a), Fig 6 (b), Fig. 6 (c), but was negatively correlated with dry matter (Fig. 7 (a) and 7 (b), Table 2). Periphyton EE content did not influence ( $p = 0.687$ ) fish growth rate (Table 2).



(a)



(b)

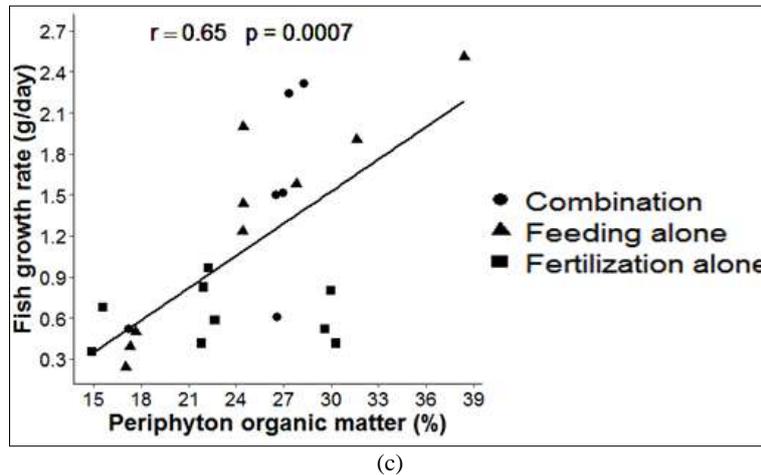


Fig 6: Relationship between *O. niloticus* growth rate and periphyton quantity (a) Biomass (b) Dry matter and (c) Organic matter

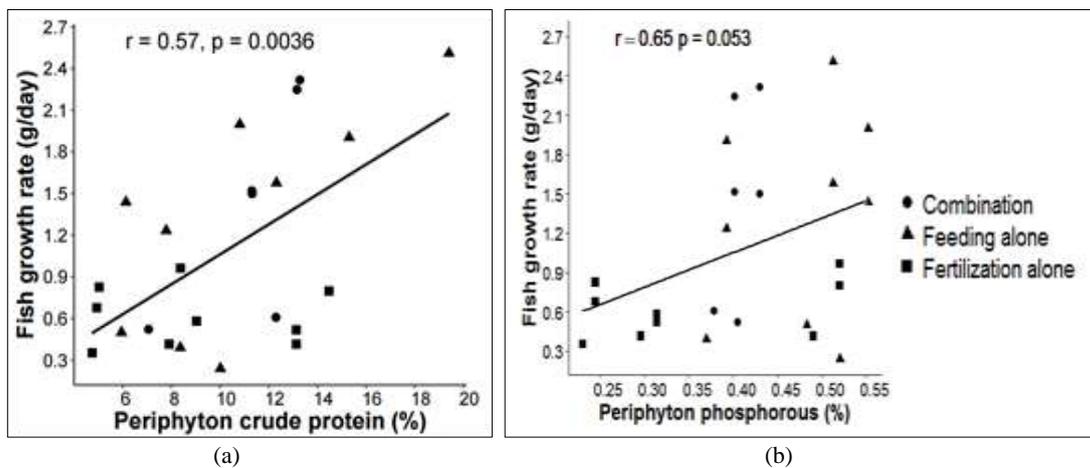


Fig 7: Relationship between *O. niloticus* growth rate and periphyton quality (a) Crude protein (b) Phosphorous

**3.5 Fish body proximate chemical composition**

Results for proximate chemical composition of Nile tilapia are shown in Table 3. Fish body proximate chemical composition values differed significantly ( $p < 0.05$ ) among the treatments. Fish grown in ponds under fertilization alone had significantly ( $p < 0.05$ ) lower percentage of DM ( $91.20 \pm 0.59\%$ ), OM ( $95.22 \pm 0.11\%$ ) and CP ( $66.43 \pm 0.45\%$ ) than those from ponds under feeding alone and the combination of fertilization plus supplementary feeding. However, the differences in DM (%), OM (%) and CP (%) contents of fish cultured under feeding alone and combination of fertilization plus supplementary feeding were

not significant (Table 3). Furthermore, fish body composition from fertilization alone treatment ( $T_1$ ) had significantly ( $p < 0.05$ ) higher ash ( $4.78 \pm 0.11$ ) and ether extract (EE) ( $18.33 \pm 0.19$ ) contents than the fish from the feeding alone ( $T_2$ ) and combination of fertilization plus supplementary feeding ( $T_3$ ) treatments. The percentage of ash content in fish carcasses did not differ ( $p > 0.05$ ) between the fish cultured under feeding alone ( $3.58 \pm 0.23$ ) and combination of fertilization plus supplementary feeding ( $3.35 \pm 0.16$ ), but EE (%) content differed significantly between fish under feeding alone and the combination of fertilization plus supplementary feeding (Table 3).

**Table 3:** Proximate chemical composition (Means  $\pm$  se) of *O. niloticus* muscles cultured in earthen ponds under fertilization alone, feeding alone and the combination of fertilization plus supplementary feeding\*

Proximate composition	Treatments		
	Fertilization alone ( $T_1$ )	Feeding alone ( $T_2$ )	Combination( $T_3$ )
DM (%)	$91.20 \pm 0.59^b$	$93.31 \pm 0.75^a$	$94.70 \pm 0.27^a$
CP (%)	$66.43 \pm 0.45^b$	$68.22 \pm 0.35^a$	$69.14 \pm 0.33^a$
OM (%)	$95.22 \pm 0.11^b$	$96.42 \pm 0.23^a$	$96.65 \pm 0.16^a$
Ash (%)	$4.78 \pm 0.11^a$	$3.58 \pm 0.23^b$	$3.35 \pm 0.16^b$
EE (%)	$18.33 \pm 0.19^a$	$16.10 \pm 0.04^b$	$14.47 \pm 0.25^c$

Means with the same superscript letter in the same row are not significantly different ( $p > 0.05$ ), Note: DM=Dry matter, CP=Crude protein, EE=Ether Extract, se=standard error.

**3.6 Influence of periphyton chemical composition on fish body chemical composition**

Results shown in Table 1 and 3 indicate that, periphyton crude protein and ether extract content influenced fish body

crude protein and ether extract. The increase of the periphyton chemical composition (CP and EE) led to the increase of the same chemical composition contents in the fish body.

## 4. Discussion

### 4.1 Periphyton biomass

The higher periphyton biomass observed in ponds under the combination of fertilization plus supplementary feeding than those under other treatments might be contributed by utilization of nutrients, especially phosphorous and nitrogen from both feeds and fertilizers applied in the ponds<sup>[30, 21]</sup>. In most cases phosphorous and nitrogen are limiting nutrients<sup>[9, 10]</sup>, but very important for promoting algae productivity and high algal biomass. This implies high primary productivity hence, high fish growth. The application of fertilizer together with the decomposition of uneaten feeds and fish excreta in the combination, promotes periphyton growth, hence, high biomass production<sup>[23, 31, 29]</sup>. Periphyton species community composition and abundance

### 4.2 Periphyton species community composition and abundance

The highest periphyton abundance were recorded in ponds that received both fertilizers and supplementary feed, suggesting that the availability of additional nutrients from the decomposition of leftover feeds might have contributed to the high abundance of plankton<sup>[32, 16]</sup>. Ponds subjected to the treatment of combination of fertilization plus supplementary feeding had higher abundance of Cyanophyceae and Zygnematophyceae than the ponds under the treatment of either feeding alone or fertilization alone. This perhaps was because of the nutrients enrichment from both fertilizer application and decomposed leftover feeds. This is in agreement with the findings elsewhere<sup>[33, 16]</sup> who found that, pond manuring increases the abundance of Cyanophyceae and total phytoplankton abundance than feeding alone.

Furthermore, the high abundance of Cyanophyceae in all treatments in the current study could be due to its tolerance to a wide range of environmental conditions and because of its toxicity. Studies have indicated that fish feed selectively on other algae rather than cyanobacteria<sup>[34, 35, 36]</sup>. However, other studies have shown that Nile tilapia has the capability to select Cyanophyceae as food in addition to Bacillariophyceae, Chlorophyceae and Euglenophyceae<sup>[2]</sup>. In the present study, ponds under feeding alone had less abundance of phytoplankton than the ponds under fertilization. It is well known that, more diversity and higher abundance of phytoplankton is found in the fertilized ponds compared to unfertilized ponds.

Pond fertilization enhances the availability of natural food for fish growth<sup>[21, 37]</sup>. The high abundance of zooplankton in the ponds under feeding alone and the combination of fertilization plus supplementary feeding was probably contributed by the presence of uneaten feeds in the pond water<sup>[2]</sup>. Other researchers have shown that the presence of organic fertilizer increases the availability of zooplankton<sup>[38, 37]</sup>. It has been reported that pond water containing the combination of uneaten feeds and inorganic fertilizer has higher zooplankton abundance than those with uneaten feeds or applied with fertilizer alone<sup>[16]</sup>.

### 4.3 Periphyton proximate chemical composition

Higher periphyton crude protein (CP %) and organic matter (OM %) contents observed in the ponds subjected to the combination of fertilization plus supplementary feeding probably was influenced by the high abundance of zooplankton observed in those ponds. Previous studies have

shown that zooplankton are rich in protein while phytoplankton contains more lipid<sup>[15]</sup>. The periphyton CP content obtained in this study was within the acceptable range from 9 to 32% CP<sup>[29]</sup> while the OM content was lower than the range between 46 and 60% reported by other studies<sup>[39]</sup>. The lower periphyton OM content (but higher ash contents) observed in the present study might be caused by high sand content in the pond water. During the experimental period, the pond water had a lot of suspended particles, especially during the rainy seasons which, in turn might have attached to the nets, and were collected together with periphyton during sampling.

Low dry matter (DM %) content for periphyton collected from the ponds under the combination of fertilization plus feeding compared to those collected from other treatments may be due to differences in species composition among the treatments. The lower periphyton DM content may also suggest high moisture content of those periphyton<sup>[40]</sup>. The higher EE content for the periphyton from the ponds under fertilization alone than those from other treatments may be caused by more light penetration due to low turbidity. This is because Secchi disk reading value was high in the ponds under fertilization alone, implying that light penetration was high<sup>[23]</sup>. Light promotes photosynthesis, thereby increases lipid content of the algae as an energy source. Also, lipid content can be influenced by pond culture condition (i.e. water quality parameters)<sup>[23]</sup>. Lipid or ether extract content in the present study was slightly lower than the value of 2 - 5% observed in marine algae<sup>[41]</sup> and extremely lower than the value of 18% which has been reported in aquaculture periphyton composition<sup>[23]</sup>. This variability might be contributed by differences in species abundance and environmental among the treatments<sup>[23, 42]</sup>.

Periphyton from the ponds under fertilization alone had lower phosphorous content than those from the other treatments probably, was due to less uptake of phosphorous from the surrounding. The availability of nutrients in the algal cell may be influenced by the quantity of nitrogen (N) and phosphorous (P) in the surrounding environment<sup>[43]</sup>. The source of nutrient in the ponds under fertilization alone was only fertilizer, whereas in ponds under the combination of fertilization plus feeding, nutrients were contributed by both fertilizer and the left-over feeds present in the ponds.

### 4.4 Relationships between periphyton quantity, quality and fish growth rate

Results in the present study show positive and significant relationships between fish growth and periphyton quantity (organic matter and biomass) and quality (crude protein content). The strong positive relationships between periphyton biomass, CP and OM contents with fish growth rate suggests that biomass, CP and OM are important factors for fish growth. This is because CP is a crucial nutrient responsible for growth and as the percentage of CP increases the growth of the fish increases<sup>[40]</sup>. Studies have shown that fish growth and body composition is reliant on feed composition and availability<sup>[44, 7, 45, 46]</sup>. Moreover, it has been shown that high OM in the periphyton constitutes important nutrients for fish growth<sup>[40]</sup>. The increase in fish growth rate as periphyton phosphorous content increased may be due to the availability of both artificial feeds and/or fertilizer which increased the amount of nutrients (including phosphorus) which is important for algae growth and hence, fish growth. It is well known that fish growth rate is influenced by the

amount of phosphorous present in the feed [7, 45,46]. Presence of phosphorous in the fish muscles enables the formation of energy (ATP) for growth and carrying out different body activities [47, 43].

#### 4.5 Fish body proximate chemical composition

Crude protein and fat contents are the major nutrients which are used to define the nutritional status of the fish [48]. Crude protein (CP %) content was higher in fish cultured in ponds under feeding alone and the combination of fertilization plus feeding. The high CP content found in the carcasses of fish cultured in ponds subjected to the treatments which involved feeding of diet is consisted with the findings of previous studies. Previous studies reported that when fish consume more supplementary feeds their body crude protein content also increases because the formulated feed which is offered to the fish has high CP content [1, 21]. The crude protein content of the feed used in this study was in the range of 25 - 30% CP which is ideal for tilapia. Also the crude protein contents (CP %) of the fish carcasses for all three treatments fall within the range reported in previous studies elsewhere [49, 24]. Fish body lipid or ether extract (EE) content significantly differed among the treatments, with the highest value being observed in fish cultured in ponds under fertilization alone. The high lipid content in the fish carcasses may be due to the high consumption of phytoplankton and zooplankton. Studies have shown that high consumption of natural food (plankton) may results into high lipid content in the fish muscles [1, 21]. The range of the fat content values observed in the present study is similar to the values of fat content which have been obtained in Nile tilapia cultured under pond system [50, 51].

Fish raised in ponds under fertilization alone had higher ash content than the fish in other treatments. The ash content of the fish body may be influenced by the type of the feed used and minerals availability in the water column [52, 24]. In the current experiment fish reared under fertilization alone solely depended on natural food available in the ponds, which probably consisted of higher proportion of minerals, especially during the rain seasons where most of the ponds had a lot of suspended organic matter. Ash content of the *O. niloticus* flesh obtained in this study falls within the range of 1.76 - 3.83% reported for tilapia species [53, 51].

#### 4.6 Influence of periphyton chemical composition on fish body chemical composition

Results have shown that, periphyton chemical composition (EE and CP contents) influenced fish body chemical composition (EE and CP contents) since there was a positive correlation between periphyton and fish chemical compositions. Previous studies have shown that fish growth and body biochemical composition may be influenced by chemical composition of the feed fed to the fish [1, 21]. In addition, it is reported that the body of fish which consume natural food accumulates more fat while those fed supplementary feed accumulates more crude protein [1, 21].

#### 5. Conclusions and Recommendations

From the results, it can be concluded that, the ponds under combination of weekly fertilization plus supplementary feeding using half of the required feed (2.5% of fish body weight) had the highest periphyton quantity (biomass, organic matter), quality (crude protein) and species abundance, which promoted high fish growth rate. The study

also revealed a positive relationship between fish body proximate chemical composition and periphyton proximate chemical composition.

It is recommended that further study should be done in different locations and seasons and essential amino acid and fatty acid composition of both periphyton and fish should be analyzed.

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