ORIGINAL RESEARCH ARTICLE

Effect of dietary supplementation with seaweed on growth and nutritional quality of Nile tilapia

***Rachel Mwendwa*[1], *Michael Wawire*[1], *Peter Kahenya*[1]**

[1]*Department of Food Science and Technology, Jomo Kenyatta University of Agriculture and Technology (JKUAT), Nairobi, Kenya.*

Corresponding author: ramwa.24@gmail.com

ABSTRACT

Feed is a major component of production costs in aquaculture, accounting for about 80% of the production costs. High-quality aquafeeds are a prerequisite to healthy and nutritious fish. Aquafeeds are expensive owing to the fact that fish oil and fish meals are the main sources of lipid and protein components, respectively. Having alternative, cheap sources of lipids in the feeds is therefore important. The brown seaweed (*Sargassum portieranum*) that is locally available on the Kenyan coast is known to be rich in omega-3 fatty acids. The objective of this study was therefore to determine the suitability of brown seaweed dietary supplementation and its effect on the nutritional quality and growth performance of Nile tilapia (*Oreochromis niloticus*).

A total of 180 male Nile tilapia fingerlings were divided into three experimental groups in triplicate. The fish were assigned to one of the three treatment diets: 0% (control diet), 5%, or 10% inclusion of the brown seaweed, and fed for 12 weeks. The weight and length (from head to tail) of the fish were measured every two weeks to determine the growth performance. At the end of the experiment, the fish muscle protein, lipid, and mineral content were determined using AOAC methods.

Seaweed supplementation significantly ($P < 0.05$) improved the body weight, length, survival, and specific growth rate of the fish, with the 10% inclusion showing higher performance than the 5%. The protein, mineral, and lipid contents of the fish muscles were also significantly affected by the seaweed supplementation. Fish fed on the 10% diet had the highest total lipid content in the muscle, at 0.93%, compared to 0.78% in the fish fed on the control diet. The protein content in the fish muscle was not significantly affected ($P < 0.05$) by the inclusion of seaweed in the feed. Overall, the results showed that supplementing the feed with 5% or 10% brown seaweed improved the growth performance and nutritional quality of tilapia fish. Thus, including brown seaweed meal in the diet of tilapia fish could offer an effective means to boost production in aquaculture.

Key words: seaweeds, Nile tilapia, aquaculture, nutritional, polyunsaturated fatty acid, growth performance

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1.0 Introduction

Fish and fish products play an important role in the human diet and health. They are sources of high-quality protein, omega-3 polyunsaturated fatty acids (*n*-3 PUFA), and micronutrients such as vitamin D, selenium, calcium, iodine, and iron (Kawarazuka & Béné, 2011; Weichselbaum *et al*., 2013). Regular consumption of fish is associated with several health benefits, such as improved neural development in infants and a reduced risk of cardiovascular inflammatory disease and insulin resistance (Chowdhury *et al*., 2012; Corella & Ordovas, 2012).

With the increase in human population and the emergence of a large number of people with greater purchasing power and a preference for animal protein over plant protein, demand for fish is increasing (Jennings et al., 2016; Kharas, 2010). Fish is supplied from two main sources: wild-capture fisheries and aquaculture. In 2018, aquaculture contributed 46% (82.1 million t) of the global fish production, of which 52% was used as food for human consumption (FAO, 2020). In the same year, the total fisheries production in Kenya was at 147,000 metric tons, with a per capita consumption of about 5 kg compared to the global consumption of 20 kg per capita (KNBS, 2020). Kenya's fish production is a major factor influencing its fish consumption; that is, an increase (or decrease) in domestic fish production tends to increase (or decrease) per capita fish consumption (Obiero *et al*., 2019). Aquaculture has great potential for growth to meet the growing demand for fish. To ensure sustainability and optimise aquaculture, all dynamics involved in production, such as feed ingredients and quality, nutrient cycling, and retention in the fish, need to be researched and understood.

Aquafeeds play an important role in aquaculture production. They account for about 50% of the variable production costs (Rana *et al*., 2009). Due to the high inclusion rate of nutrients like protein (up to 40%) in feeds for fingerlings, the cost of feeds is relatively high (Cho *et al*., 2003). In addition, fish oil is the source of polyunsaturated fatty acids (PUFA) in aquafeeds but is expensive (Klinger & Naylor, 2012). Approximately 75% of annual fish-oil production is used as a feed ingredient in aquafeeds (Auchterlonie, 2018). This in turn reduces the amount of fish oil available for human consumption, and this is exacerbated by the stagnation in the production of fish oil (Shepherd & Jackson, 2013). These factors combined to necessitate the need for sustainable and cheap alternative sources of PUFA to use in aquafeeds. Seaweeds are a promising feed ingredient since they are a source of the omega-3 fatty acids eicosapentaenoic acid (EPA) and docosahexaenoic acid (DHA) (FAO, 2020). Omega-3 fatty acids are important in fish for cellular metabolism and maintenance of cell membrane structure and integrity (Miller *et al*., 2008; Tocher, 2010).

Seaweeds are a rich source of carbohydrates, protein (with a high content of essential amino acids), and minerals like magnesium, calcium, iodine, and sodium (Bocanegra *et al*., 2009; Fleurence, 1999; Miyashita *et al*., 2013). The brown seaweeds (*Phaeophyta*) have been reported to have the highest lipid content among the seaweeds, with predominantly long-chain PUFA, eicosapentaenoic acid (EPA, C20:5 n-3), and arachidonic acid (ARA, C20:4 n-6) (Wan *et al*., 2019). Although seaweeds may not have as much lipid content as other plant

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sources, the proportion and quality of their fatty acids are high in polyunsaturated fatty acids (PUFA). In some seaweed species, PUFA can account for up to 40% of total fatty acids (Nomura *et al.,* 2013).

Several studies have demonstrated that dietary supplementation with seaweeds in a fish diet can greatly improve the muscle lipid profile, especially the omega-3 fatty acids (Garcia-Vaquero & Hayes, 2016; Güroy *et al*., 2013). A study by Dantagnan *et al*. (2009) reported an increase in muscle total PUFA and omega-3 PUFA of up to 73% and 64%, respectively, in rainbow trout when fed on the brown seaweed, *Macrocystis pyrifera.* Furthermore, the inclusion of plant-based ingredients at various levels in feeds affects the final product quality (lipid, amino acid, color, and texture) (De Francesco et al., 2004).

In assessing the suitability of a feed ingredient for use in aquafeed, determining its nutrient utilisation is one key step (Glencross, 2020). This involves feeding trials and then assessing the growth responses. Therefore, the objective of this study was to evaluate the effect of supplementation with brown seaweed (*Sargassum portieranum*) on the growth performance, muscle biochemical composition, and lipid profile of Nile tilapia (*Oreochromis niloticus*).

2.0 Materials and methods

2.1 Experimental site

The research study was carried out at the ViclnAqua aquaculture hatcheries in Kisumu, Kenya. All the laboratory analyses were conducted at Jomo Kenyatta University of Agriculture and Technology in the Food Biochemistry Laboratory.

2.2 Feed formulation and proximate composition

Forty kilogrammes of seaweed (*Sargassum portieranum)* were collected from Shimoni, on the south coast of Kenya, in the month of July. The seaweeds were hand-picked and then washed with seawater to remove foreign particles. The samples were then transported to the Jomo Kenyatta University of Agriculture and Technology for cold storage (4^{o}C). They were thoroughly washed in running tap water, dried in a conventional hot-air oven at 40^{o}C for 24 hours, and then ground into a fine powder. The other feed ingredients, i.e., fish meal, shrimp, wheat bran, cassava flour, and vegetable oil, were purchased from local stores.

For the diet preparation, the dry base ingredients were ground in a mill and then sieved through a 1 mm sieve mesh. The ingredients were then weighed out in triplicate and homogenised for preparing the experimental diets. The proportions of ingredients used are as shown in Table 1. Fishmeal was used at 10% in each diet, as recommended in organic aquaculture. The seaweed was added to two diets at inclusion levels of 5% and 10%, with a control diet without seaweed at 0%. Sunflower oil and water were added and thoroughly mixed to make a blend of soft dough consistency. The dough was extruded using an automated meat mincer fitted with a 2 mm plate. The 2-mm pellets were sundried to a constant weight. Airtight containers were used for storage of the pellets until the start of the feeding.

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2.3 Experimental set-up

Male Nile tilapia fingerlings were obtained from the experimental site (ViclnAqua) hatcheries. One hundred and eighty fingerlings were distributed randomly in 9 circular cylindrical tanks (3 replicates per treatment) at a stocking density of 20 fingerlings per tank. The fingerlings began with an average weight of 31.11 ± 0.60 g and 12.5 ± 0.04 cm length (head to tail). Feeding was done to apparent satiation, three times daily (at 0830, 1300, and 1700 hours) for 12 weeks.

The fish were maintained at a natural photoperiod. Dissolved oxygen was maintained above 6 mg/l using an aeration system. The quality of the water was regulated by replacing the water in the tanks three times every week. At the end of the experimental period, the fish were fasted for 24 hours and then sampled.

Table 1: Feed formulation and proximate composition of the diets. Ingredients content in the feed are in g/kg.

Ingredients	Diets (g/kg, dry weight basis)			
	Diet 1 (0%, control)	Diet 2 (5%)	Diet 3 (10%)	
Fish meal	100	100	100	
Shrimp (*Caridina nilotica*)	152	152	152	
Wheat bran	222	222	222	
Wheat pollard	222	222	222	
Sunflower meal cake	169	163	163	
Cassava flour (binder)	75	75	75	
Vegetable oil	50	50	50	
S. portieranum	0	50	100	
[a] Mineral and vitamin premixes	10	10	10	
[b] Proximate composition (% dry matter basis)	Diet 1	Diet 2	Diet 3	P value
Dry matter	95.86±0.18[a]	94.68±0.03[b]	94.92±0.13[b]	0.005
Crude protein	30.96±0.27[b]	32.76±0.67[ab]	34.90±0.81[a]	0.048
Crude lipids	5.57±0.22[a]	5.76±0.21[a]	5.55±0.08[a]	0.683
Crude ash	6.53±0.27[b]	7.18±0.56[b]	8.75±0.24[a]	0.016
Crude fibre	10.83±0.38[a]	10.67±0.21[a]	10.48±0.21[a]	0.682
[c] NFE	41.92±0.18[a]	37.82±0.03[ab]	35.62±0.13[b]	0.053

Diet 1, 0% inclusion; Diet 2, 5% inclusion; Diet 3, 10% inclusion of the seaweed.

[a] Vitamin and mineral premix composition per Kg of feed: Vitamin A, 600 I.U.; vitamin D3, 100 I.U.; vitamin E, 3 I.U.; vitamin K (menadione), 0.42 mg; vitamin B1, 0.25 mg; vitamin B2, 0.6 mg; vitamin B6, 0.5 mg; vitamin B12, 0.0011 mg; nicotinic acid, 2.5 mg; pantothenic acid, 2.2 mg; folic acid, 0.15 mg; biotin, 0.001 mg; vitamin C, 1 mg; copper, 0.5 mg; manganese, 15 mg;

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zinc, 4.5 mg; iodide, 0.14 mg; selenium, 0.012 mg; cobalt, 0.02 mg; choline chloride, 15 mg; iron 4 mg.

[b] Proximate values are the mean ± S.E.

[c] Nitrogen-free extract (NFE) = 100 - (content of moisture + crude protein + crude lipids + crude ash + fiber)

2.4 Analytical methods

2.4.1 Growth performance

The growth performance of the fish was determined by measuring its weight, length, survival rate, and food index parameters (specific growth rate, condition factor, and feed conversion ratio). The parameters were calculated using equations from Tekinay & Davies, 2001, as follows:

$$\boldsymbol{Survival\ rate\ (SR,\%) = \frac{Number\ of\ live\ tilapia\ at\ end\ of\ experiment}{Intial\ number\ of\ tilapia} \times 100} \quad \text{..... Eq. 1}$$

$$\boldsymbol{Weight\ gain\ (WG,g) = Average\ final\ weight - Average\ initial\ weight} \quad \text{..... Eq. 2}$$

$$\boldsymbol{Length\ gain,(LG,cm) = Average\ final\ length - Average\ initial\ length} \quad \text{..... Eq. 3}$$

$$\boldsymbol{Specific\ growth\ rate\ (SGR,\%) = \frac{(Log\ of\ final\ weight(g) - Log\ of\ initial\ weight\ (g))}{Experiment\ period} \times 100} \quad \text{... .... Eq. 4}$$

$$\boldsymbol{Condition\ factor\ (CF) = \frac{Final\ weight\ (g)}{Final\ lenght\ (cm^3)}} \quad \text{..... Eq. 5}$$

2.4.2 Proximate analysis

Moisture content, crude protein, crude lipid, crude fat, crude fiber, and ash for experimental diets and fish muscle were determined according to Association of Official Analytical Chemists method specification 950.46 (AOAC, 1995). The moisture content was determined by weighing 2 g of the sample into a moisture dish and transferring it to an oven previously heated to temperatures of 105 °C, where it was dried for 3 hours. The final weight of the sample was taken after the drying period and cooling in a desiccator. The loss in weight was reported as moisture content (AOAC, 1995, method 925. 10). Ash content was determined by incineration of the samples in an Advantec KL-420 electric muffle furnace at 550 °C for 12 hours. The crude protein content (N × 6.25) was determined by the semi-micro Kjeldahl method after acid digestion using a Kjeldahl system (Velp scientifica model). The Kjeldahl system was used to digest 5 g of the sample mixed with two catalysts (5 g of K_2SO_4 and 0.5 g $CuSO_4$) and 15 ml of concentrated H_2SO_4. The digest was then distilled and finally titrated to obtain the nitrogen content. The crude protein was obtained by multiplying the nitrogen content by the protein factor. Crude lipid was analysed using the Soxhlet system (Geohardt model). About 5 g of the sample was weighed into thimbles, and lipid extraction was done using petroleum ether in a soxhlet apparatus for 8 hours. The extraction solvent was evaporated, and the remaining lipid was dried in an oven at 70 °C to a constant weight to obtain the crude lipid. The crude fibre content was determined using the Hennenberg-Stohmann method (AOAC, 1995), which

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involves sequential digestion of samples with 1.25% H_2SO_4 and 1.25% NaOH, followed by drying at 105 ^oC for 30 min and ashing at 550 ^oC for 1 hour, and then cooling.

2.4.3 Fatty acid profiling

The Bligh and Dyer method (1959) protocol was used to extract lipid from fish muscle. Finely ground samples were homogenised using a methanol-chloroform (2:1, v/v) mixture containing 0.01% butylated hydroxytoluene (BHT), and the extract was filtered with Whatman No. 1 filter paper. A second solvent mixture of methanol, chloroform, and water (2:1:0.8) was added to the extract, and the process was repeated. The mixture was then centrifuged at 3000 rpm for 10 minutes, and the chloroform layer at the bottom was separated from the aqueous layer using a micropipette. The chloroform layer was transferred into a reflux flask and evaporated to dryness using a rotary vacuum evaporator. Five (5) ml of methanolic H_2SO_4 (1% H_2SO_4, v/v) was added to the extract, and esterification was done at 70 ^oC for 3 hours. The fatty acid methyl esters (FAME) were then extracted into 5 mL hexane and 100 mL water. The mixture was transferred into a separating funnel, and the hexane layer (bottom) was withdrawn and passed through anhydrous sodium sulphate. The extract was finally dried to 0.5 ml using the rotary vacuum evaporator. Concentrated FAME extract was then transferred to vials. Gas chromatography-mass spectrophotometry (Agilent Technologies, model 7890B) was used to identify the FAMEs by injecting the FAME extract into a silica capillary column ((SUPELCO, Omegawax^{tm} 530). The injection temperature and detection temperature were 240 ^oC and 260 ^oC respectively.

2.5 Statistical analysis

All the experimental data are expressed as the mean ± standard error (SE). Data was subjected to one-way analysis of variance (ANOVA), followed by the Duncan multiple-range test to compare differences among treatments. When $P < 0.05$, statistically significant differences between means were considered. Statistical analysis was performed using R version 4.0.2 software.

3.0 Results

3.1 Growth performance

The results on the growth performance and survival of Nile tilapia are presented in Table 2. The final weight and weight gain of the fish increased with the increasing levels of seaweed supplementation and weight gain of the fish increased with the increasing levels of seaweed supplementation. The fish fed on the diet containing 10% seaweed had the highest final body weight (66.12 ± 2.24 g) and weight gain (28.08 ± 1.03 g). The three diets had no significant variation on the final length or the gain in length of the fish. There was variation in the survival rate and specific growth rate after 12 weeks of feeding on the unsupplemented diet and the supplemented diet at a 10% inclusion level. The survival rate and specific growth rate of the experimental fish were significantly influenced by the different levels of seaweed supplementation. The specific growth rate of the fish increased with the increase in the level of seaweed used in supplementation.

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Table 2: Growth performance and survival rate of Nile tilapia fed for 12 weeks on three different diets

Diets	Weight (g)		Length (cm)		SR (%)	SGR (% per day)	CF
	Final	gain	Final	gain			
Diet 1	59.19±1.03^{b}	28.08±1.03^{b}	15.63±0.29^{a}	3.13±0.29^{a}	93.33±3.33^{ab}	0.33±0.01^{b}	0.02±0.00^{a}
Diet 2	62.61±1.54^{ab}	31.50±1.54^{ab}	15.62±0.48^{a}	3.12±0.48^{a}	100±0.00^{a}	0.36±0.01^{ab}	0.02±0.00^{a}
Diet 3	66.12±2.24^{a}	35.00±2.24^{a}	16.37±0.26^{a}	3.87±0.26^{a}	100±0.00^{a}	0.38±0.02^{a}	0.02±0.01^{a}
LSD	5.797	5.797	1.230	1.230	6.660	0.047	0.002

The values are the ± SE of the means. n = 3. The values in the same column with different superscript letters differ significantly (P < 0.05). Abbreviations: SGR (specific growth rate); SR (survival rate); CF (condition factor). Diet 1, 0% inclusion; Diet 2, 5% inclusion; and Diet 3, 10% inclusion of seaweed

3.2 Proximate analysis

As shown in Table 3, the crude fibre and crude lipid of the fish were significantly (P < 0.05) increased when fed diets supplemented with seaweeds compared to the control (0%). Fish fed on the 10% supplemented diet showed the highest crude lipid and fibre content at 0.93% and 0.39%, respectively. The three diets showed no significant difference in the dry matter or crude protein levels in the fish muscle. The results of crude ash content showed no significant difference (P < 0.05) between the unsupplemented diet and the diet supplemented with 10% seaweed.

Table 3: Proximate composition of the fish muscle

% Proximate composition (wet weight basis)	Dry matter	Crude protein	Crude lipid	Crude ash	Crude fibre
Diet 1	21.76±0.21^{a}	19.26±0.17^{a}	0.78±0.07^{b}	1.13±0.02^{a}	0.23±0.02^{b}
Diet 2	22.17±0.38^{a}	20.02±0.27^{a}	0.91±0.05^{a}	0.88±0.08^{b}	0.36±0.03^{a}
Diet 3	22.52±0.42^{a}	20.38±0.54^{a}	0.93±0.05^{a}	1.18±0.02^{a}	0.39±0.02^{a}
LSD	1.451	1.261	0.111	0.159	0.081

Data are expressed as the mean ± SE; n = 3. The superscript letters in the same column differ significantly (p<0.05). Diet 1, 0% inclusion; Diet 2, 5% inclusion; and Diet 3, 10% inclusion of the seaweed

3.3 Fatty acid profiling

At the end of the experimental period, fish supplemented with seaweeds showed no significant difference in the saturated fatty acids (palmitic and stearic acid) and monounsaturated fatty acids (palmitoleic), except for the oleic acid, which significantly (P<0.05) increased in fish fed with a diet of 5% inclusion. Seaweed supplementation at two levels (5% and 10%) significantly increased omega-3 content while decreasing omega-6

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content. For the three dietary treatments, linoleic acid was the most abundant fatty acid in the fish muscle. The diet with 5% seaweed supplementation showed the highest omega-3/omega-6 ratio. The total omega-6 levels were significantly higher than the other unsaturated fatty acid groups (MUFA and omega-3) for the three dietary treatments, as shown in Figure 1.

Table 4: Fatty acid profile of the fish muscle fed on three different diets

% Fatty acids	Diets			
	Diet 1	Diet 2	Diet 3	P-Value
SFA				
C16:0	21.91±0.02^{a}	20.33±0.44^{a}	21.23±0.90^{a}	0.223
C18:0	9.41±0.59^{a}	12.11±0.93^{a}	9.88±0.78^{a}	0.103
MUFA				
C16:1	2.30±0.26^{a}	3.08±0.40^{a}	1.85±0.69^{a}	0.215
C18:1	2.33±0.39^{b}	10.82±0.65^{a}	1.45±0.33^{b}	0.000
Omega 6				
C18:2	37.34±0.43^{a}	24.65±0.83^{c}	32.06±0.16^{b}	0.001
C18:3	0.20±0.02^{b}	0.44±0.02^{a}	0.29±0.03^{b}	0.005
C20:4	6.37±0.67^{b}	7.47±0.63^{ab}	8.59±0.33^{a}	0.085
Omega 3				
C20:5 n-3	1.44±0.09^{b}	2.21±0.35^{ab}	2.39±0.21^{a}	0.065
C22:6 n-3	19.51±0.38^{b}	21.97±0.42^{b}	26.16±0.91^{a}	0.011
Totals				
SFA	30.86±0.68^{a}	32.44±0.55^{a}	31.90±0.93^{a}	0.536
MUFA	4.64±0.71^{b}	13.73±0.02^{a}	3.31±0.34^{b}	0.000
LC-PUFA, ω-6	43.26±0.56^{a}	31.94±0.60^{c}	40.61±0.02^{b}	0.000
LC-PUFA, ω-3	21.04±0.35^{c}	23.93±0.03^{b}	28.54±0.55^{a}	0.002
ω-3/ ω-6 (ratio)	0.49±0.01^{b}	0.75±0.01^{a}	0.70±0.01^{b}	0.002

The values are the ± SE of the means. n = 3. The values in the same row with different superscript letters differ significantly ($P < 0.05$). Diet 1, 0% inclusion; Diet 2, 5% inclusion; and Diet 3, 10% inclusion. Abbreviations: SFA, saturated fatty acid; MUFA, monounsaturated fatty acid; PUFA, polyunsaturated fatty acid; LC-PUFA, long-chain polyunsaturated fatty acid; C16:0, palmitic acid; C18:0, stearic acid; C16:1, palmitoleic acid; C18:1, oleic acid; C18:2, linoleic acid; C20:4, arachidonic acid; C18:3, linolenic acid; C20:5, eicosapentaenoic acid; C22:6, docosahexaenoic acid; ω-6, Omega 6; ω-3, Omega 3.

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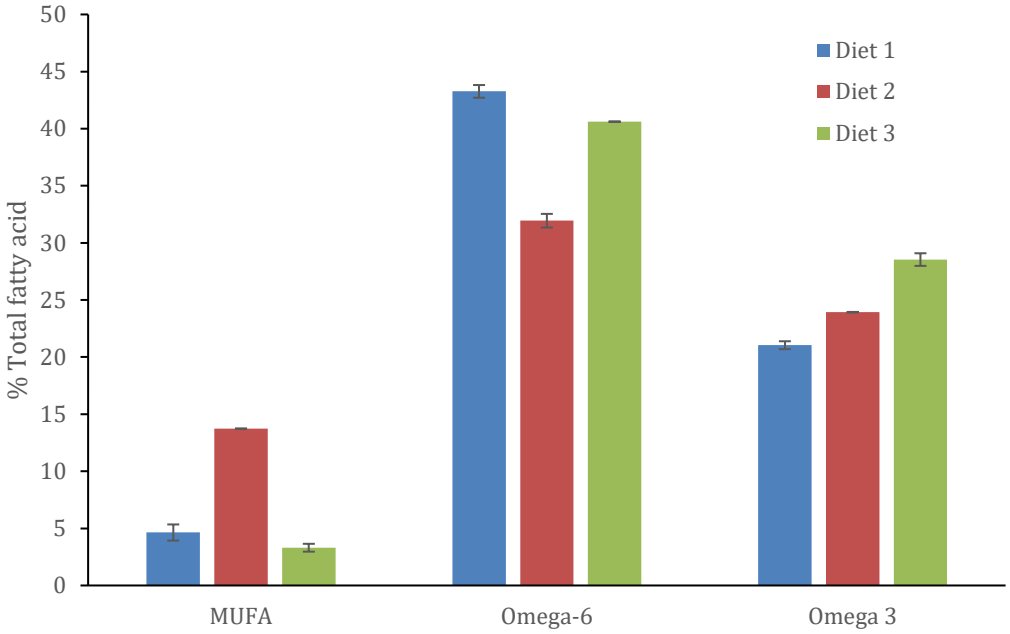

Figure 1: Comparison of the unsaturated fatty acid in the fish muscle fed three different diets for 12 weeks. Diet 1, 0% inclusion; Diet 2, 5% inclusion; Diet 3, 10% inclusion

4.0 Discussions

Previous studies have demonstrated that inclusion of small amounts of seaweeds in fish diets could enhance the growth performance of several cultured fish species (Nakagawa & Montgomery, 2007; Roy *et al*., 2011), including *Sargassum* (Ragaza *et al*., 2013; Serrano *et al.,* 2015). In the present study, supplementation of fish feed with *S. portieranum* showed improved fish growth, indicating that inclusion of seaweed in tilapia feeds can improve weight gain without greatly affecting other growth performance parameters. A study by Ergün *et al.* (2008) showed similar results when *Ulva sp*. was included at a rate of 5% in the Nile tilapia diet. The current study is also consistent with those of Wassef *et al.,* (2013) and Khalafalla & El-Hais (2015), who found that diets containing seaweeds improved fish growth, feed efficiency, and hematology. Furthermore, the performance of rainbow trout was enhanced when they were fed diets supplemented with 0.4% of brown seaweed (Ribeiro *et al.,* 2017). On the contrary, previous experiments on different fish species fed different diets supplemented with different levels of brown seaweed meals did not have a significant impact on growth performance. However, differences that arise in trials have been attributed to the species of seaweed, fish species, and type of feed, water quality, age, weight, or level of inclusion (Abdelrhman *et al.,* 2022).

The survival rate of the fish fed the unsupplemented diet was relatively low compared to the supplemented diets as a result of the higher mortality rate of the fish. The higher survival rate in the supplemented diet could be attributed to the immunoactivity of seaweeds since the rearing conditions (water quality) were similar in all the treatments.

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The positive effect could also be attributed to the bioactive phytochemical molecules found in seaweeds, which have been shown to improve immune responses and growth performance (Van Doan et al., 2017), as well as the presence of sulphate carbohydrates in seaweeds (Fernández et al., 2010, Fernández et al., 2011), which trigger nonspecific immunity, making them a major amplifier to immunity and growth accelerators (Telles *et al.,* 2018). Furthermore, including seaweeds and their extracts in the diets of aquatic animals improves immunity and growth parameters (Sharawy et al., 2020).

The specific growth rate was comparable in the 5% and 10% supplementation groups, with both outperforming the control diet. Earlier studies had demonstrated that high inclusion levels (>10%) have a detrimental effect on fish growth or provide no additional benefit to the fish (Azaza *et al.,* 2008; El-Tawil, 2010; Güroy *et al*., 2007). Moreover, recommendations for the maximum inclusion level in Nile tilapia have been established at 10% for the green seaweed *Ulva sp.* and 5% for the red seaweed *Gracilaria sp.* (Marinho *et al*., 2013; Silva *et al*., 2015). Thiessen *et al*. (2004) demonstrated that growth performance could be aggravated when using seaweeds in fish diets since plant ingredients constitute a certain amount of fibre that may be detrimental to their nutritional value and palatability.

Analysis of the body composition is an effective measure of the health and physiological condition of fish (Saliu *et al.,* 2007); further, it is vital in optimising their utilisation (Martin *et al*., 2000). The proximate components, lipid and fibre, in the fish muscle were positively affected by the increase in the incorporation of seaweed in the feeds, except for the protein and ash content, which were similar to the control. This increase suggests that seaweeds could have influenced the absorption and synthesis of the two nutrients in Nile tilapia. Additionally, Nile tilapias have high amylase activity and hence prioritise carbohydrates as an energy source and spare proteins (Kamunde *et al*., 2019). Indeed, several studies on the utilisation of seaweed as a feed ingredient have demonstrated that the assimilation of nutrients in fish is dependent on the level of inclusion and species of both the seaweed and fish (Peixoto *et al*., 2016; Valente *et al*., 2006; Wan *et al*., 2019).

The nutritional profile of fish, especially the fatty acid profile, is often a reflection of its diet (Bell *et al*., 2003; Rosenlund *et al*., 2002; Keriko *et al*., 2021). Therefore, feeding a diet rich in PUFA shows high levels of PUFA in the muscle of various fish species (Li *et al.,* 2013; Tonial *et al*., 2009; Visentainer *et al*., 2005). In this study, the results showed a similar pattern: the PUFA content in the fish muscles that were fed the supplemented diets improved with the increase in inclusion levels. These results are similar to what EL-Tawil (2010) found for red tilapia fed a diet with the green seaweed *Ulva sp.* at 10% inclusion. The omega-3 fatty acid levels were increased by about 14% and 36% in the fish feed diets supplemented with 5% and 10%, respectively. These results could be attributed to an increase in the quantity of seaweed incorporated into the diets. All three diets resulted in fish tissue with higher docosahexaenoic acid (DHA) levels than eicosapentaenoic acid (EPA), with the fish fed the diet supplemented with 10% seaweed showing the highest levels of DHA. PUFAs play an important role in the growth and survival of marine fish (Tocher, 2010). These processes utilise more EPA,

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conserving more DHA relative to the EPA, hence resulting in high DHA levels in the fish muscle (Rønnestad *et al.,* 1995; Villalta *et al*., 2005). Moreover, omega-6 levels were relatively high in all the fish fed the two supplemented experimental diets, with linoleic acid contributing the highest levels. The omega-3/omega-6 ratio increased with the increase in the seaweed proportion in the diet, suggesting active metabolism of the essential fatty acid from the dietary source (Lim *et al*., 2009).

5.0 Conclusions

From the findings in this study, incorporation of 5% and 10% of the brown seaweed *Sargassum portieranum* in aquafeeds improves the fatty acid content of Nile tilapia while at the same time promoting its growth performance and proximate composition. Thus, including brown seaweed at low levels in fish feeds has the potential to improve the polyunsaturated fatty acid content of fish while enhancing growth.

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6.2 Conflict of interest

None.

6.3 General acknowledgment

Authors of this manuscript contributed equally.

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