

Article



Evaluation of *Sargassum* spp. Meal Inclusion in Diets for Prejuvenile Nile Tilapia

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Abstract: This preliminary study evaluated dietary inclusion of *Sargassum* spp. meal in feeds for prejuvenile Nile tilapia (*Oreochromis niloticus*) through method-specific feed characterization and a 67-day feeding trial. Two experimental diets containing 10% and 15% *Sargassum* spp. meal were compared with a commercial reference feed using 54 fish distributed among nine aquaria, with three independent aquaria per treatment and six fish per aquarium. The *Sargassum* spp. meal contained 4.24 ± 0.19 g/100 g moisture and 37.41 ± 0.91 g/100 g ash on the as-analyzed gravimetric basis. NIRS-based feed protein estimates were 52.62 ± 0.15 , 53.35 ± 0.31 , and 54.44 ± 0.46 g/100 g of scanned material for the 10%, 15%, and commercial diets, respectively. Gravimetric and NIRS outputs were generated from separate aliquots and method-specific reporting bases and were therefore not interpreted as a common wet-weight mass balance. At day 67, aquarium-level final body weight was 59.90 ± 12.31 g, 68.56 ± 3.83 g, and 57.15 ± 4.33 g in the 10%, 15%, and commercial treatments, respectively. One-way ANOVA based on aquarium means detected no treatment effect on final body weight [$F(2,6) = 1.724$, $p = 0.256$] or final total length [$F(2,6) = 1.597$, $p = 0.278$]. The 15% diet showed numerically higher weight gain and SGR and a lower FCR, but confidence intervals were wide, and the differences were not statistically significant. These results support only the preliminary short-term feasibility of the tested formulations; they do not establish equivalence, superiority, complete nutritional adequacy, or consumer safety. Confirmation requires greater experimental replication and characterization of digestibility, amino acids, minerals, iodine, arsenic, antinutritional factors, and tissue bioaccumulation.

Keywords: *Sargassum* spp.; *Oreochromis niloticus*; aquafeed; macroalgae; feed formulation; growth performance; aquarium-level analysis; preliminary feeding trial

1. Introduction

Aquaculture has become one of the most important animal-food sectors for global nutrition, and recent FAO reporting shows that farmed aquatic animals now account for a larger share of global aquatic animal production for human consumption than capture fisheries. Within this context, Nile tilapia (*Oreochromis*



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niloticus) remains one of the most relevant freshwater species because of its rapid growth, high consumer acceptance, tolerance to diverse farming conditions, and importance for food security in tropical and subtropical regions (FAO, 2024; El-Sayed, 2006; National Research Council [NRC], 2011). These traits make tilapia especially attractive for low- and medium-input production systems, but they also place strong pressure on aquafeed formulation because feed quality strongly influences early growth, uniformity, and production efficiency (El-Sayed, 2006; NRC, 2011).

Feed remains the highest operating cost in many finfish systems, and the long-standing reliance on conventional ingredients such as fishmeal and fish oil continues to raise economic and environmental concerns. Although the aquafeed industry has progressively reduced marine ingredient inclusion rates, these commodities still play strategic nutritional roles, especially as concentrated sources of digestible protein, essential amino acids, and long-chain lipids (Tacon & Metian, 2015; NRC, 2011). In tropical aquaculture, this creates a practical need for partial replacement strategies using locally available, lower-cost, and more sustainable ingredients that complement rather than abruptly displace conventional protein sources (FAO, 2024; Tacon & Metian, 2015; El-Sayed, 2006). In tilapia farming, this search is especially relevant because the species is often promoted as a food-security fish, so feed innovation must balance nutritional adequacy, affordability, and regional availability (FAO, 2024; El-Sayed, 2006).

One biomass that has gained unusual relevance in the tropical Atlantic and Caribbean is pelagic *Sargassum*. Since 2011, massive recurrent blooms associated with the Great Atlantic *Sargassum* Belt have affected coastlines from West Africa to the Caribbean and Gulf of Mexico, turning an open-ocean habitat-forming macroalga into a major coastal management problem when it reaches shore in excessive volumes (Wang et al., 2019; Robledo et al., 2021; Rodríguez-Martínez et al., 2025). Once stranded, decomposing *Sargassum* can reduce light penetration, alter coastal water quality, promote hypoxic conditions, and damage seagrass and coral-associated habitats; it also creates odor, cleanup, and economic burdens for coastal communities and tourism-dependent regions (van Tussenbroek et al., 2017; Robledo et al., 2021; Rodríguez-Martínez et al., 2025). Consequently, recent literature increasingly frames *Sargassum* not only as an environmental nuisance but also as a candidate for valorization into fertilizers, biomaterials, bioenergy, food-related products, and feed ingredients (Robledo et al., 2021; Rodríguez-Martínez et al., 2025).

Macroalgae comprise green, red, and brown groups with different biochemical profiles. *Sargassum* is a genus of brown macroalgae; here, *Sargassum* spp. denotes the collected biomass identified at genus level, without a species-resolved composition. It does not denote an undifferentiated mixture of green, red, and brown algae. Brown macroalgae are typically low in fat but may provide minerals, structural polysaccharides, dietary fiber, and bioactive compounds. Their protein content varies with species, season, habitat, and processing (Holdt & Kraan, 2011; Mæhre et al., 2014; Cherry et al., 2019; Peñalver et al., 2020; O’Sullivan et al., 2010). We therefore evaluate *Sargassum* meal as a complementary feed ingredient rather than a complete protein source.

Previous feeding studies demonstrate that responses to macroalgal meals depend on algal identity, inclusion level, processing, and diet formulation. In Nile tilapia, 10% *Ulva* meal maintained growth comparable with a control diet, whereas 15% and 20% inclusion reduced growth and increased FCR (Marinho et al., 2013). Low-level *Ascophyllum nodosum* meal did not alter final weight or length and improved apparent FCR in a 42-day fingerling trial (Costa et al., 2013). Conversely, replacing 20% or more of fishmeal with *Gracilaria arcuata* reduced growth, with the adverse response increasing at higher inclusion levels (Younis et al., 2018). Digestibility studies also show that tilapia can utilize nutrients from several macroalgae, including *Sargassum*, while emphasizing species-specific variation (Pereira et al., 2012). These findings support cautious evaluation of moderate inclusion levels rather than assuming a uniform benefit across macroalgae.

This study evaluated whether two diets containing 10% or 15% locally collected *Sargassum* spp. meal affected growth and feed conversion in prejuvenile Nile tilapia (*Oreochromis niloticus*) relative to a commercial reference over 67 days. We tested the hypothesis that growth performance and feed conversion differed among the three dietary formulations; the null hypothesis was equality of treatment means. Specifically, we compared aquarium-level weight and length gains, specific growth rate, and feed conversion ratio; characterized the algal meal, feeds, and muscle material using the available analytical methods; and quantified Cd, Pb, and Hg in the experimental feeds. These comparisons focus on complete formulations because other ingredients also differed between diets.

2. Materials and Methods

2.1. *Sargassum* Meal Preparation and Experimental Diet Formulation

Sargassum spp. biomass was collected from the Caribbean coast of Colón, Panama, and processed into meal. Briefly, the collected biomass was cleaned to remove algal residues, animal remains, and visible impurities; washed with 0.9% saline solution; sun-dried for five days; milled; sieved to 63 μm ; packaged; subjected to proximal analyses; and stored until diet formulation. Figure 1 shows the processing workflow.

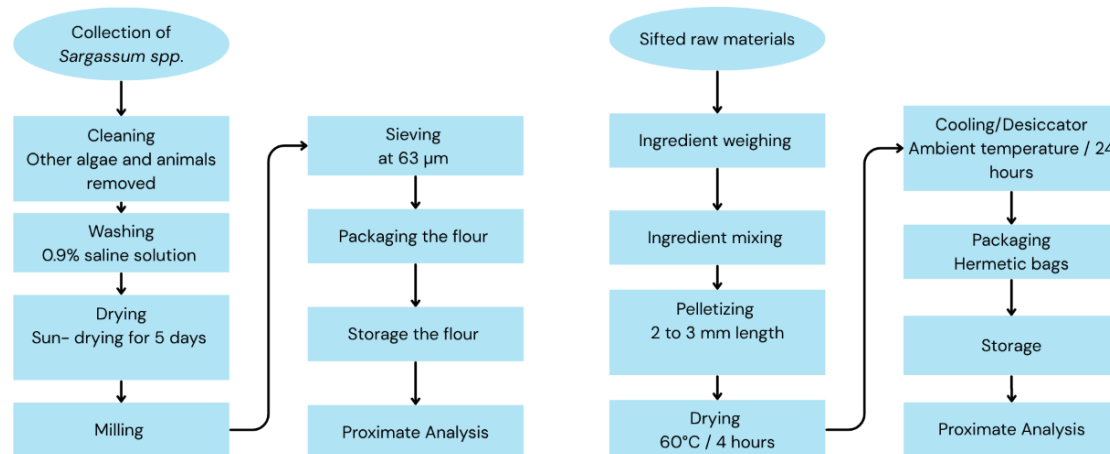


Figure 1. Methodological flowchart for production of *Sargassum* spp. meal and preparation of experimental pelleted feed. In the original workflow, ambient temperature referred only to the 24-h post-drying cooling period inside a desiccator, not to aquarium-water temperature.

Two experimental diets containing *Sargassum* spp. meal were prepared. Diet A contained 10% *Sargassum* meal, and Diet B contained 15%. The quantitative formulation records show that increasing *Sargassum* inclusion coincided with reductions in fishmeal and corn and an increase in gluten; therefore, *Sargassum* inclusion was not the only formulation difference between Diets A and B. Both experimental diets were intended to provide approximately 45% protein and were compared with Inicio Cromis (BioMar), a commercial reference feed. The full formulation of the commercial feed was unavailable. Table 1 summarizes the available formulation details.

Table 1. Formulation records for the experimental diets and identification of the commercial reference feed.

Ingredient	Diet A, 10% <i>Sargassum</i> Meal	Diet B, 15% <i>Sargassum</i> Meal
Corn	0.97	0.12
Soybean meal	0.10	0.08
Gluten	6.00	13.70
Fishmeal	69.00	57.97
<i>Sargassum</i> meal	8.50	12.70
Corn oil	6.00	6.00
Fish oil	6.00	6.00
Carboxymethyl cellulose (CMC)	1.60	1.60
Yeast	1.20	1.20
Baking powder	0.12	0.12
Vitamin-mineral premix	0.51	0.51
Target crude protein (%)	Target: 45	Target: 45

Note: Ingredient quantities are expressed as g per 100 g of complete formulated feed. The nominal 10% and 15% *Sargassum* meal inclusion levels refer to the flour fraction of the experimental formulations (84.57 g), corresponding to 8.50% and 12.70% of the complete diets, respectively.

2.2. Feeding Trial and Analytical Determinations

We conducted a 67-day feeding trial with 54 prejuvenile Nile tilapia (*Oreochromis niloticus*). Fish were allocated to Diet A (10% *Sargassum* meal), Diet B (15% *Sargassum* meal), or the commercial reference feed using nine aquaria, three independent aquaria per treatment, and six fish per aquarium (Figure 2). The aquaria were continuously aerated. We offered feed in three daily rations at approximately 07:00, 12:00,

and 16:00, and adjusted ration amounts weekly based on measured biomass. Biometric measurements were recorded weekly, and operational water-quality checks were recorded twice weekly. The aquarium, not the individual fish, was the independent experimental unit; fish measurements within each aquarium were summarized before inference. The archived records did not retain sufficiently complete numerical series or descriptions for aquarium volume, stocking density by volume, water source, water-exchange rate, aquarium-cleaning procedure, photoperiod, temperature, dissolved oxygen, pH, ammonia, nitrite, or nitrate. These parameters were therefore not reconstructed retrospectively, and the incomplete husbandry record is acknowledged as a study limitation.

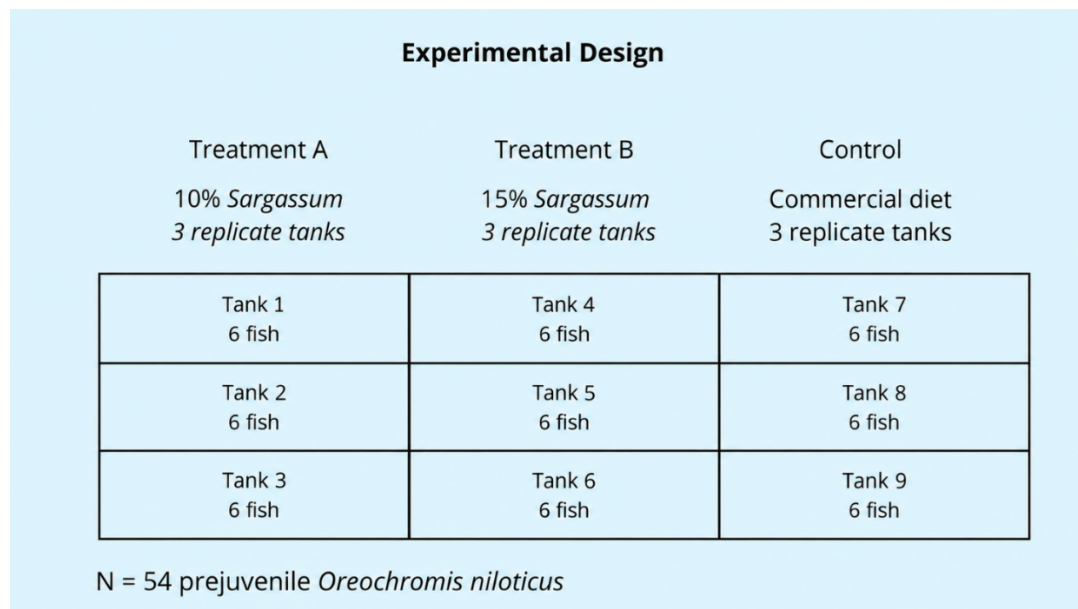


Figure 2. Experimental layout of the 67-day feeding trial with prejuvenile Nile tilapia (*Oreochromis niloticus*). The trial included three dietary treatments: 10% *Sargassum* spp. meal, 15% *Sargassum* spp. meal, and commercial feed, with three replicate aquaria per treatment and six fish per aquarium, for a total of 54 fish.

We characterized *Sargassum* meal, feeds, and muscle tissue using method-specific procedures (Table 2). Moisture and ash were determined gravimetrically on separate aliquots using AOAC 925.10:2005 and AOAC 923.03:2005 (AOAC International, 2005). Feed protein and fat were estimated by near-infrared spectroscopy (NIRS™ DS2500 F; FOSS Analytical A/S, Hillerød, Denmark) using the feed calibration. For muscle analysis, scales, organs, and fins were removed; muscle material was dried at 103.2 °C for 3 h, cooled in a desiccator, pulverized, and scanned using the fish-tissue calibration. The reported muscle moisture and ash values originated from separate gravimetric aliquots, whereas protein and lipid values were instrument outputs from dehydrated, pulverized material. Consequently, gravimetric and NIRS columns are method-specific analytical outputs and do not constitute a common wet-weight mass balance and should not be summed.

Table 2. Analytical methods and reporting basis used for *Sargassum* meal, feeds, muscle material, and growth outcomes.

Sample Type	Variables Analyzed	Method/Equipment
<i>Sargassum</i> meal	Moisture, ash, pH, titratable acidity	AOAC 925.10:2005; AOAC 923.03:2005; AOAC 943.02:2005; AOAC 939.05
Experimental and commercial feeds	Moisture and ash (gravimetric); protein and fat (NIRS); pH; acidity; Cd, Pb, and Hg for experimental feeds only	AOAC methods; NIR™ DS2500 F; FAAS for Cd and Pb; CV-AAS for Hg
Tilapia muscle tissue	Moisture and ash from gravimetric aliquots; protein and lipids from dried, pulverized material by NIRS	AOAC methods and NIRS
Growth performance	Aquarium-level weight, length, SGR, and FCR	Weekly biometrics and Excel processing

2.3. Determination of Cadmium (Cd), Lead (Pb), and Mercury (Hg)

We analyzed the two experimental feed formulations in duplicate. We digested each replicate by microwave-assisted acid digestion following Method 3051A (U.S. Environmental Protection Agency [EPA], 2007), adapted for food matrices. Approximately 0.5 g of homogenized sample was weighed into PTFE digestion vessels and mixed with HNO₃:HCl: ultrapure H₂O (6:3:6, v/v/v). Digestion was performed using a QWave microwave system (Questron Technologies Corp., Mississauga, Canada) at 130 °C for 10 min, 150 °C for 5 min, and 170 °C for 5 min. After cooling, the solutions were diluted to 25 mL, filtered, and stored until analysis. Cd and Pb were determined by FAAS (Thermo Scientific iCE 3000 Series; Thermo Fisher Scientific, Cambridge, United Kingdom) according to AOAC 999.10, and Hg by CV-AAS using a RA-4500 analyzer (Nippon Instruments Corporation, Takatsuki, Japan). We used calibration curves, reagent blanks, duplicates, and spiked samples for quality control. Only the two experimental diets had validated archived results; we did not reconstruct or infer commercial-feed, original-meal, or fish-tissue metal concentrations. Instrumental operating conditions are summarized in Table 3.

Table 3. Instrumental operating conditions and analytical parameters for the determination of Cd, Pb, and Hg by atomic absorption spectrometry.

Parameter	Cadmium (Cd)	Lead (Pb)	Mercury (Hg)
Technique	FAAS	FAAS	CV-AAS
Equipment	Thermo Scientific iCE 3000	Thermo Scientific iCE 3000	Nippon RA-4500
Wavelength (nm)	228.8	217.0	253.7
Method limit of quantification (LOQ)	0.10 mg/kg	2.5 mg/kg	0.5625 µg/kg
Calibration curve R ²	0.9992	0.9987	0.9999

2.4. Data processing and Statistical Analysis

We recovered weekly source data separately for all nine aquaria. We calculated growth variables for each aquarium, yielding three independent observations per dietary treatment. Final body weight, final total length, weight gain, length gain, SGR, and FCR were summarized as aquarium-level mean ± standard deviation with two-sided 95% confidence intervals based on the *t* distribution. We compared treatments using one-way ANOVA with 2 and 6 degrees of freedom. Omega-squared (ω^2) was reported as an exploratory effect-size estimate, and negative estimates were truncated at zero. We independently verified statistical calculations from the aquarium-level source values in Python 3.12 using SciPy 1.17. Given the small number of independent aquaria, we treated all inferential analyses as exploratory; we did not interpret nonsignificant findings as equivalence, non-inferiority, or the absence of a biologically meaningful effect. Analytical triplicates used for method-specific feed characterization were treated as technical analytical replicates and were not interpreted as independent feeding-trial units.

$$\text{SGR (\% day}^{-1}\text{)} = 100 \times [\ln(\text{FBW}) - \ln(\text{IBW})] / t \quad (1)$$

$$\text{FCR} = \text{Feed offered (g)} / \text{Weight gain (g)} \quad (2)$$

where FBW is the final body weight (g), IBW is the initial body weight (g), Feed offered is the total feed offered during the feeding trial (g), and *t* is the duration of the experimental period (days).

3. Results

3.1. Method-Specific Characterization of Sargassum Meal, Feeds, and Muscle Material

Gravimetric analysis of *Sargassum* spp. meal yielded 4.24 ± 0.19 g/100 g moisture and 37.41 ± 0.91 g/100 g ash on the as-analyzed basis, together with pH 6.86 ± 0.05 and titratable acidity $0.052 \pm 0.010\%$. These values describe the processed material for the variables measured; protein, lipid, amino acids, fiber, carbohydrates, energy, iodine, arsenic, and antinutritional compounds were not determined in the algal meal. Table 4 summarizes the method-specific analytical results.

The NIRS feed scans produced protein estimates of 52.62 ± 0.15 , 53.35 ± 0.31 , and 54.44 ± 0.46 g/100 g scanned material for the 10%, 15%, and commercial diets, respectively. The 15% formulation had lower gravimetric ash and lower NIRS fat than the 10% formulation. This pattern is not attributed to *Sargassum* alone because the 15% diet simultaneously contained less fishmeal and corn and more gluten, and the study did not use a one-factor ingredient-replacement design. Moisture, ash, NIRS protein, and NIRS fat differed

among the three feeds in technical-replicate comparisons, whereas pH did not; these method-level comparisons do not overcome the limited biological replication of the feeding trial.

Table 4. Method-specific analytical outputs for *Sargassum* spp. meal, feeds, and dehydrated tilapia muscle material.

Matrix	Sample	Moisture (Gravimetric, as Analyzed; g/100 g)	Ash (Gravimetric, as Analyzed; g/100 g)	Lipid/Fat (NIRS Output; g/100 g Scanned Material)	Protein (NIRS Output; g/100 g Scanned Material)	pH
Algal meal	<i>Sargassum</i> spp. meal	4.24 ± 0.19	37.41 ± 0.91	ND	ND	6.86 ± 0.05
Feed	10% <i>Sargassum</i> diet	19.21 ± 0.20	46.17 ± 0.22	12.12 ± 0.13	52.62 ± 0.15	6.29 ± 0.06
Feed	15% <i>Sargassum</i> diet	17.67 ± 0.15	41.26 ± 0.35	10.72 ± 0.26	53.35 ± 0.31	6.24 ± 0.07
Feed	Commercial feed	22.24 ± 0.22	55.26 ± 0.15	13.71 ± 0.19	54.44 ± 0.46	6.19 ± 0.02
Muscle tissue	10% <i>Sargassum</i> diet	21.79 ± 0.30	5.64 ± 0.28	12.25 ± 0.05	52.59 ± 0.01	ND
Muscle tissue	15% <i>Sargassum</i> diet	20.85 ± 0.23	5.29 ± 0.43	10.83 ± 0.04	53.13 ± 0.04	ND
Muscle tissue	Commercial feed	17.79 ± 0.09	4.44 ± 0.57	13.76 ± 0.04	54.95 ± 0.14	ND

Note: Values are mean ± SD. Gravimetric moisture, ash, and pH were based on three analytical replicates. Feed NIRS estimates were based on three scans; available muscle NIRS estimates were based on two scans. ND = not determined. Gravimetric and NIRS outputs were obtained from separate aliquots and method-specific reporting bases and must not be summed as a common wet-weight composition. Muscle material was dehydrated, cooled, and pulverized before NIRS and therefore does not represent fresh muscle on a wet-weight basis.

3.2. Heavy-Metal Screening of Experimental Feeds

Cd, Pb, and Hg were quantified in the two experimental feeds as a preliminary feed-level screening (Table 5). Concentrations were above the analytical LOQ and are reported descriptively. Because the archived dataset did not contain corresponding measurements for the commercial feed, original *Sargassum* meal, or fish tissues, the results cannot be used to compare contaminant transfer among ingredients or treatments, assess bioaccumulation, or establish consumer safety.

Table 5. Cadmium, lead, and mercury concentrations measured in the two experimental feeds.

Treatment	Cd (mg/kg dw)	Pb (mg/kg dw)	Hg (µg/kg dw)	Interpretation
10% <i>Sargassum</i> diet	0.256 ± 0.00021	2.60 ± 0.00014	0.89 ± 0.000707	Feed-level screening only
15% <i>Sargassum</i> diet	0.269 ± 0.00035	2.79 ± 0.0015	0.98 ± 0.0014	Feed-level screening only
Method LOQ	0.10	2.50	0.5625	—

Note: dw = dry weight. Values are means of two analytical replicates (n = 2). These measurements do not include arsenic, iodine, broader mineral profiling, the commercial feed, the original algal meal, or fish tissues. No comprehensive regulatory or consumer-safety conclusion was drawn from this restricted three-metal feed screen.

3.3. Growth Performance of Prejuvenile Nile Tilapia

All nine aquaria showed progressive increases in mean body weight and total length over 67 days (Figure 3). Aquarium-level final body weight was 59.90 ± 12.31 g in the 10% treatment, 68.56 ± 3.83 g in the 15% treatment, and 57.15 ± 4.33 g in the commercial treatment. Corresponding final total lengths were 14.05 ± 0.99, 14.92 ± 0.09, and 14.36 ± 0.31 cm. The 15% formulation had numerically higher final weight, weight gain, length gain, and SGR and a lower FCR; however, these descriptive differences were based on three aquaria per treatment, and their confidence intervals overlapped broadly (Table 6).

Table 6. Aquarium-level growth outcomes after 67 days, reported as mean ± SD with 95% confidence intervals.

Treatment	Final Weight (g)	Weight Gain (g)	Final Length (cm)	Length Gain (cm)	SGR (% day ⁻¹)	FCR
10% <i>Sargassum</i>	59.90 ± 12.31 [29.32, 90.49]	42.93 ± 12.56 [11.73, 74.13]	14.05 ± 0.99 [11.58, 16.52]	4.38 ± 1.04 [1.79, 6.97]	1.86 ± 0.35 [1.00, 2.72]	1.87 ± 0.37 [0.95, 2.79]
15% <i>Sargassum</i>	68.56 ± 3.83 [59.03, 78.08]	51.32 ± 3.14 [43.52, 59.11]	14.92 ± 0.09 [14.71, 15.13]	5.46 ± 0.15 [5.09, 5.84]	2.06 ± 0.07 [1.88, 2.24]	1.43 ± 0.06 [1.29, 1.58]
Commercial	57.15 ± 4.33 [46.40, 67.89]	40.19 ± 4.06 [30.12, 50.27]	14.36 ± 0.31 [13.58, 15.14]	4.69 ± 0.29 [3.97, 5.42]	1.81 ± 0.09 [1.58, 2.04]	1.78 ± 0.16 [1.38, 2.19]

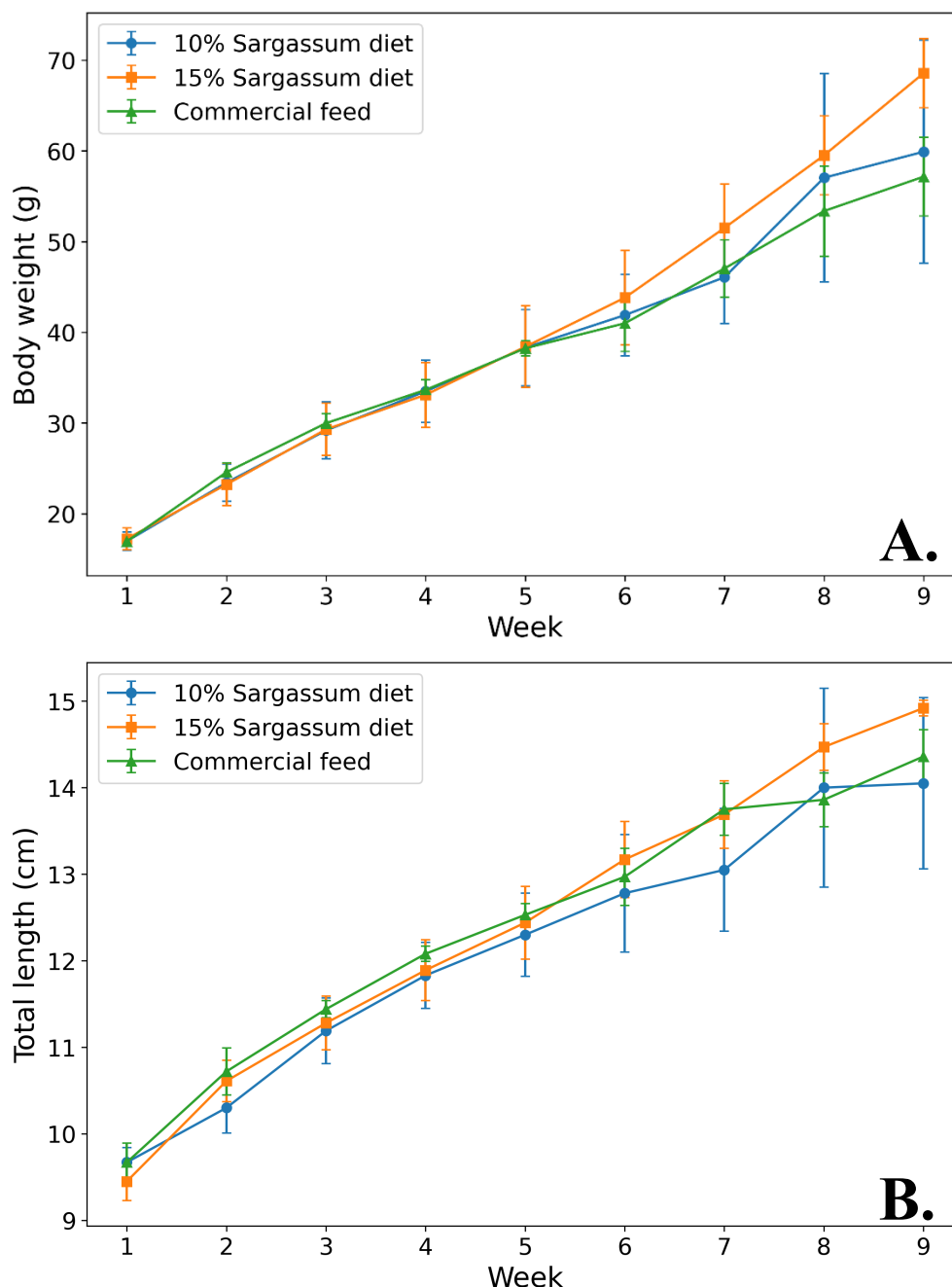


Figure 3. Weekly growth performance of prejuvenile Nile tilapia (*Oreochromis niloticus*) fed diets containing 10% and 15% *Sargassum* spp. meal and a commercial reference feed during a 67-day feeding trial. (A) Mean body weight progression. (B) Mean total length progression. Values are expressed as aquarium-level means $\pm$ standard deviation from three independent replicate aquaria per treatment ($n = 3$).

The 95% confidence intervals are wide, particularly for the 10% treatment, and reflect the limited number of independent aquaria. The numerical ranking of treatment means therefore remains hypothesis-generating and is not evidence that the 15% diet was superior or that either experimental diet was equivalent to the commercial reference.

3.4. Aquarium-Level Statistical Synthesis of Growth Outcomes

Initial aquarium means did not differ detectably among treatments for body weight [$F(2,6) = 0.093$, $p = 0.913$] or total length [$F(2,6) = 1.091$, $p = 0.394$]. Aquarium-level ANOVA detected no statistically significant treatment effect for final weight, weight gain, final length, length gain, SGR, or FCR (Table 7). The largest exploratory ω^2 estimates were observed for FCR (0.290) and length gain (0.231), but their p values remained above 0.05 and the small sample produced substantial uncertainty.

Table 7. One-way ANOVA and exploratory omega-squared effect sizes based on three independent aquaria per treatment.

Outcome	F(2,6)	p	ω^2
Initial weight	0.093	0.913	0.000
Final weight	1.724	0.256	0.139
Weight gain	1.643	0.270	0.125
Initial length	1.091	0.394	0.020
Final length	1.597	0.278	0.117
Length gain	2.348	0.176	0.231
SGR	1.177	0.370	0.038
FCR	2.841	0.136	0.290

Treatment comparisons used aquarium means, with three independent observations per diet. Weekly measurements describe growth trajectories within the same experimental units and do not increase the number of independent replicates. The nonsignificant tests and wide confidence intervals therefore warrant cautious interpretation.

4. Discussion

All three formulations supported growth during the 67-day trial, but the aquarium-level analysis did not detect differences in growth or feed conversion. The hypothesis of different treatment means was therefore not supported at the 0.05 significance level. The 15% diet yielded a higher mean final weight and a lower mean FCR than the other diets (Table 6), which warrants further evaluation of that formulation. However, three aquaria per treatment provide limited precision, and the wide confidence intervals leave biologically relevant differences unresolved. These results establish a preliminary feeding response to the tested diets, not an optimal *Sargassum* inclusion level.

Comparisons with other Nile tilapia studies show why both algal identity and dietary level matter. Marinho et al. (2013) fed juvenile *Oreochromis niloticus* diets containing 0%, 10%, 15%, or 20% *Ulva* spp. meal for 63 days. Growth at 10% was similar to the control, whereas growth declined at 15% and 20% and FCR increased with inclusion. Our 15% *Sargassum* formulation did not show that pattern, but this does not establish that *Sargassum* performs better than *Ulva*: the algal material, diet composition, and rearing conditions differed. In a 12-week trial with the same fish species, Güroy et al. (2007) tested *Ulva rigida* and *Cystoseira barbata* separately at 5%, 10%, and 15% dietary inclusion. The 15% *Ulva* diet produced the poorest growth and FCR, whereas the brown alga did not show the same growth penalty. Together, these studies indicate that a threshold observed for one macroalga cannot be transferred directly to another.

Lower supplementation levels address a different nutritional question. Costa et al. (2013) fed Nile tilapia (*Oreochromis niloticus*) fingerlings *Ascophyllum nodosum* meal at 0, 5, 10, 15, or 20 g/kg feed, equivalent to 0%, 0.5%, 1%, 1.5%, or 2%, for 42 days. Final weight and length did not differ among treatments, while apparent FCR improved with increasing supplementation. Those levels are well below the 10% and 15% used here. The result supports evaluating brown algae as supplements, but cannot establish nutritional adequacy when algae constitute a larger fraction of the diet. Fish also began that trial at approximately 0.43 g, compared with about 17 g here, limiting direct comparison of absolute growth and feed conversion.

Fishmeal replacement percentages must also be distinguished from whole-diet inclusion. Younis et al. (2018) tested *Gracilaria arcuata* in Nile tilapia (*Oreochromis niloticus*) at 20%, 40%, and 60% replacement of fishmeal over 12 weeks. Their formulation table corresponds to 7%, 14%, and 21% algal meal in the complete diets. Growth was lower than in the control at all three levels, although FCR did not differ significantly between the control and the lowest replacement treatment. Thus, the reported 20% treatment cannot be interpreted as 20% dietary algae or used as a universal inclusion limit. In African catfish (*Clarias gariepinus*), Al-Asgah et al. (2016) tested the same red alga at fishmeal replacement levels of 10%, 20%, and 30%, corresponding to 4.5%, 9%, and 13.5% of the complete diets. Growth and feed utilization were similar to the control at the lowest level and poorer at the higher levels. These comparisons illustrate variation among fish species and formulations; they do not identify a transferable *Sargassum* threshold.

Digestibility provides a more direct link between ingredient composition and nutrient utilization than growth alone. Pereira et al. (2012) tested diets containing 30% *Porphyra dioica*, *Ulva* spp., *Gracilaria vermiculophylla*, or *Sargassum muticum* in Nile tilapia (*Oreochromis niloticus*) and rainbow trout (*Oncorhynchus mykiss*). In tilapia, *Sargassum* and *Ulva* had the highest calculated protein digestibility among

the tested seaweeds, but digestibility of the seaweed-containing diets was generally below that of the reference diet. This distinction matters: comparatively favorable digestibility among algae does not establish equivalence to conventional feed ingredients. Their identified *S. muticum* also differs from our genus-level Caribbean biomass. Without digestibility or amino-acid measurements here, the observed growth cannot be attributed to improved protein availability or a specific algal constituent.

The measured ash content of the algal meal, 37.41 g/100 g on the as-analyzed basis, is relevant to formulation because a large mineral fraction leaves less material available to supply organic nutrients. Seaweed composition varies with species and processing (Mæhre et al., 2014), and high ash and structural carbohydrate contents can constrain its use in animal diets (Makkar et al., 2016). Minerals and bioactive compounds may also have functional value, but neither their availability nor their physiological effects was tested here. Moreover, the lower measured ash in Diet B, despite its higher *Sargassum* inclusion, shows that the composition of the entire mixture matters. Reductions in fishmeal and increases in gluten occurred simultaneously, so the nutrient contribution of the alga cannot be separated from those changes.

The analytical results also require comparison on compatible reporting bases. Gravimetric measurements and NIRS outputs in this study originated from separate aliquots and preparations; summing them would not yield a valid proximate composition. Likewise, NIRS values from dried muscle cannot be compared directly with fresh-muscle percentages or interpreted as nutrient retention. This limits comparisons with conventional carcass analyses in feeding studies such as Younis et al. (2018). The measured commercial-feed results remain useful as descriptive observations, but its unavailable formulation prevents an ingredient-matched comparison. Consequently, neither apparent protein concentration nor numerical growth rankings demonstrate that the experimental diets met all nutritional requirements (NRC, 2011).

The contaminant measurements address a separate feed-development requirement. Cd, Pb, and Hg were measured in the two experimental feeds. Still, their concentrations cannot be assigned to *Sargassum* because the original algal meal and other ingredients were not analyzed for these metals. Seaweed valorization must account for both nutritional composition and potential contaminants (Cherry et al., 2019; Robledo et al., 2021). Without commercial-feed and fish-tissue measurements, the present results do not establish comparative exposure, bioaccumulation, or consumer safety. Incomplete water-quality records further limit comparisons across feeding environments. The practical contribution is therefore evidence that these locally prepared formulations supported short-term tilapia growth, along with analytical information identifying the remaining constraints. Nutrient availability, standardized composition, contaminant transfer, and production costs must be resolved before commercial adoption.

5. Conclusions

In this preliminary 67-day trial, diets containing 10% or 15% *Sargassum* spp. meal supported progressive growth of prejuvenile Nile tilapia. Aquarium-level analyses found no statistically significant treatment effects on final weight, weight gain, final length, length gain, SGR, or FCR. Because only three independent aquaria were available per treatment, the findings do not establish equivalence, non-inferiority, superiority, or absence of biologically meaningful dietary effects relative to the commercial reference.

The favorable numerical means observed for the 15% formulation remain hypothesis-generating. Method-specific feed characterization, an incompletely characterized algal meal, simultaneous changes in several formulation ingredients, incomplete husbandry records, and a restricted three-metal screen limit nutritional, mechanistic, sustainability, and safety conclusions. These findings support further evaluation of the formulations under controlled nutritional and husbandry conditions before practical recommendations are made.

Author Contributions

A.L.G. contributed to the conceptualization of the study, funding acquisition, project administration, data curation, formal analysis, methodology, validation, supervision, and the writing of the original draft, as well as the review and editing of the final manuscript. D.P. contributed to the conceptualization, methodology, investigation, and supervision of the bioassay. J.B. contributed to the conceptualization, methodology, and investigation. F.R. contributed to data curation, formal analysis, methodology, and validation. F.C. contributed to conceptualization, methodology, and investigation. C.J. contributed to the methodology, investigation, and supervision. J.M. contributed to data curation, formal analysis, visualization, validation, and to the writing, review, and editing of the manuscript. L.M. contributed to the

methodology, provided laboratory and logistical resources for the study, and participated in the review of the manuscript. A.O. contributed to the conceptualization, methodology, investigation, and supervision of the bioassay. All authors have read and approved the final version of the manuscript.

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Institutional Review Board Statement

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Informed Consent Statement

This study did not involve human participants.

Data Availability Statement

The results supporting the findings are reported in this article. Requests for the underlying aquarium-level and analytical data should be directed to the corresponding author.

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Conflicts of Interest

Given their roles as Guest Editors, Ana L. García and Jay Molino were not involved in the peer review of this paper and had no access to information regarding its peer-review process. Full responsibility for the editorial process of this paper was delegated to another editor of the journal.

Use of AI and AI-Assisted Technologies

During the preparation of this work, the authors used Grammarly and ChatGPT (OpenAI) to assist with language editing. The authors reviewed and edited the content and took full responsibility for the published article.

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