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Unwinding the potential of plant-based diets in *Oreochromis niloticus*: An evaluation of growth performance and survivability

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Abstract
A study was conducted to analyze the growth performance of mixed-sex Nile Tilapia (*Oreochromis niloticus*) with soyabean-enriched feed and moringa-enriched feed. The experiment was conducted for 75 days in a cemented tank (3 m x 2 m x 1 m) in a completely randomized design with three treatments in triplicate. The treatments include: T1 - Control (Homemade feed), T2 - homemade feed + 5% soybean meal, and T3 - homemade feed + 5% Moringa leaf powder. Nile tilapia fish were added to each tank at a rate of 5 fish/m^2, with an average stocking weight of 7.7±0.3 g/fish. The fish were fed with designated feed at a rate of 3% of their body weight during the culture period. Different water quality parameters, such as water temperature, dissolved oxygen, and pH, were within the optimum range for fish culture. The average final weight was significantly higher ($p<0.05$) in T2 (27.60±0.10) and T3 (27.40±0.06) compared to T1 (26.01±0.10). Although there was no significant difference in survival rate among treatments ($p > 0.05$), the daily weight gain (DWG) was significantly greater ($p < 0.05$) in T2 (0.266 ± 0.004 g/day) and T3 (0.262 ± 0.02 g/day) compared to T1 (0.245 ± 0.003 g/day). The apparent feed conversion ratio (AFCR) was lowest in T3 (1.94 ± 0.02), indicating better feed utilization efficiency. In contrast, the feed conversion efficiency (FCE) was lowest in T1 (0.48 ± 0.012), meaning only 48% of the feed was converted into fish biomass. Thus, it can be concluded that the inclusion of plant-based ingredients such as soybean meal and moringa leaf powder in fish diets can significantly enhance growth performance and feed efficiency. Therefore, plant-based diets may serve as sustainable and effective alternatives to animal-based feeds in aquaculture.

Keywords: Nile Tilapia; Plant-based diets; Moringa leaves powder; Soyabean meal

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Introduction
Aquaculture is one of the fastest-growing subsectors of global food production, playing a vital role in food and nutrition security while helping conserve wild fish populations (Ul Hassan et al., 2021). As the global population grows and wild fisheries face challenges, such as overfishing, fish farming is expanding

worldwide to meet the increasing consumer demand for fish and related products (Dawood, 2020). The share of aquaculture in total fish production has increased from 42.2% in 2012 to 51% (FAO, 2022). Advancements in culture systems, feed technology, and management practices have supported this growth. However, improving the sustainability and cost-efficiency of aquaculture remains a pressing challenge; particularly in the area of fish nutrition (Caruso, 2015). Among production costs, feed constitutes the largest share, accounting for up to 40–60% of total expenses (Moniruzzaman & Fatema, 2022). Fish meal (FM) has traditionally been the primary protein source due to its excellent nutritional profile, including high protein content, digestibility, and essential fatty acids (Hardy & Tacon, 2002; Jackson, 2006). However, rising prices and limited availability of fish meal have prompted the search for more affordable and sustainable protein alternatives (Higgs et al., 1995; Mansour et al., 2021).

Plant-based proteins have emerged as promising substitutes, with soybean meal being one of the most widely used due to its high digestibility, availability, and cost-effectiveness (Lemos et al., 2000). Its suitability, however, varies depending on species-specific feeding behavior and size (Sánchez-Lozano et al., 2009). Previous studies have demonstrated the successful inclusion of soybean meal in the diets of several aquaculture species, including Red Drum, Atlantic Salmon, Asian Seabass, and Gilthead Sea Bream (McGoogan & Gatlin, 1997; Refstie et al., 1998; Bonaldo et al., 2008).

Another promising plant-based alternative is *Moringa oleifera*, a fast-growing, drought-resistant plant commonly found in the southern plains of Nepal. Its leaves are rich in protein, essential minerals, carotenoids, and antioxidants, with low levels of antinutritional compounds (Ferreira et al., 2008). Studies have shown its potential as a supplementary protein source in fish diets, particularly for *Labeo rohita* and other freshwater species (Chiseva, 2006; Ayotunde et al., 2011). Despite this potential, few studies have assessed moringa-based feeds in Nepalese aquaculture systems.

Therefore, this study aims to evaluate the effects of incorporating soybean meal and *Moringa oleifera* leaf powder into the diets of Nile tilapia (*Oreochromis niloticus*) reared in cement tanks in Nepal. Specifically, the study assesses feed consumption, growth performance, feed efficiency, and survival rates to determine the feasibility of using these locally available plant-based ingredients as alternatives to conventional fish meal.

Materials and Methods

Experimental site

The experiment was conducted in the Fisheries Research Unit at the Directorate of Agricultural Research (DOAR), Parwanipur, Bara, which lies in the Central Terai region of Nepal. The research farm lies at latitude 27°4'40.9" N and longitude 84°56'9.85" E, 75 meters above sea level. The culture period was 75 days from 15 September 2024 to 27 November 2024. The advanced fingerlings of Nile tilapia (mixed sex) were procured from the fish farm of the Fisheries Research Unit, Parwanipur.

Experimental setup

The experiment was conducted in a completely randomized design (CRD) with three treatments and three replications of each. Mixed-sex tilapia were stocked at a Stocking density of 5 fish/m², corresponding to 30 fish per tank, and a total of 270 fish were used in the entire experimental setup.The treatments were as follows:

Table 1: Treatment with experimental feed

Treatment No.	Treatment
T_1	Control (Home-made feed)

T_2	Home-made feed + soyabean meal (5%)
T_3	Home-made feed + moringa leaf powder (5%)

Feed preparation

Supplementary pellet feeds were prepared by traditionally used feed ingredients, viz., mustard oil cake, rice bran, to make a 25% crude protein diet. The required feed composition was calculated by the Hit-Trial method in MS Excel. In order to prepare feed, moringa leaves were plucked from the tree and sun-dried for 3-4 days and ground in a mixer to form moringa powder. MOC was soaked early night to remove toxins present in it. In the early morning of the other day, Rice bran and MOC were mixed in a 1:1 ratio to form a home-made diet (T1). Rice bran, MOC, and Soyabean meal were mixed in the right proportion to form soybean-enriched feed (T2). Similarly, Moringa powder, rice bran, and MOC were mixed in the right amount to form a moringa-enriched feed (T3). The ingredients were thoroughly mixed, and water was gradually added to the mixture until a stiff dough was formed. This dough was then pelletized using a manual pelletizer to produce uniform feed pellets. The pellets were subsequently oven-dried at 45 °C for 24 hours to reduce moisture content and ensure shelf stability. Finally, the dried pellets were stored in airtight containers to prevent spoilage and maintain quality throughout the feeding trial.

Table 2: Diet composition of experimental feed

S.N.	Ingredients	T_1	T_2	T_3
1.	Rice bran	50	50	50
2.	MOC	50	45	45
3.	Soyabean meal	-	5	-
4.	Moringa powder	-	-	5
	Total	100	100	100

Tank preparation, fish stocking, and feeding

Before the experiment, the tank was cleaned and water was filled into the tank. After completion of tank preparation, the tilapia fry were released into a 3 m* 2 m cemented tank. Seeds were procured from the fish farm of the Fishery research unit, Parwanipur, and stocked in a tank at a stocking density of 5 fish/m^2. The tilapia fry ranged from 6g to 10g in size, with an average stocking density size 7.7±0.3 g/fish). Altogether, 30 fish were stocked in each tank. The feeding rate was 3% of the body mass throughout the culture period. Feeds were in the form of pellets. Manual feeding was done twice a day, between 8-9 am and 3-4 pm.

Water quality analysis

Throughout the experiment, the dissolved oxygen level, water temperature, and pH were monitored weekly using a Lutran-DO 5519 and a Milwaukee pH meter to determine the fluctuation of water quality parameters.

Sampling of fish

Sampling of fish was done every 15 days to ensure healthy and desirable growth of fish. For this, 15 cultured fish were taken out from each tank, and their body weights were measured by using a weighing balance. Fish were released into the hapa after sampling, and feeding was not done on sampling day, as they would be in a stressful condition. The feed required up to the next sampling was adjusted from the recorded total biomass of the fish. At the end of the experiment, fish harvesting was done after 75 days with the help of a drag net. All the fish were collected, counted, and weighed to assess the survival rate.

Analytical parameters

Proximate analysis of feeds

Proximate analysis of feed was performed using the standard protocol of the Analysis of the Office Association of Official Chemists (AOAC, 1990) at the National Animal Nutrition Research Center, Khumaltar, Lalitpur. The analysis includes the moisture content, crude protein (CP), crude fiber (CF), and Ether Extract (EE) of T1 (Control), T2

(soyabean-enriched feed), and T3 (moringa-enriched feed) used in the study.

Data management and analysis

The following formulae were used to evaluate growth performance parameters, including specified growth rate (SGR), survival rate (SR), feed conversion ratio (FCR), feed conversion efficiency (FCE), daily weight gain (DWG), and average weight gain (AWG):

$$\text{DWG (g)} = \frac{\text{Mean final weight (g/fish) - Mean initial weight (g/fish)}}{\text{Total culture period (days)}}$$

$$\text{AWG (g)} = \text{final weight - Initial weight}$$

$$\text{FCR} = \frac{\text{Quantity of feed fed (g)}}{\text{Total weight gain (g)}}$$

$$\text{FCE} = \text{weight gain/total feed provided} \times 100$$

$$\text{Survival rate (\%)} = \frac{\text{Total number of fish harvested}}{\text{Total number of fish stocked}} \times 100$$

Statistical analysis

The collected data were entered into an Excel spreadsheet on Windows 2011 and presented as the mean ± standard error. One-way analysis of Variance (ANOVA) was conducted to compare the effects of different diets on growth performance. Following this, Tukey's multiple comparisons test was used to evaluate specific differences between the diets and growth performance among the treatments. Values with a significance level of $P<0.05$ were considered significant. All analyses were performed using Statistical Package for the Social Sciences (SPSS) version 30 for Windows.

Results

Proximate analysis of feeds

The proximate composition of different feeds prepared and used as different treatments in Table 3. The protein content of all feed ranged between 26.27%-30.04%. The moisture content in Treatment 3 used as moringa-enriched feed was lowest compared to other treatments while crude fiber in diets containing soyabean meal seemed to be higher than other diets.

Table 3. Proximate analysis of feed types used in control and treatment tanks in present experiment

Parameter	Control / T1	T2	T3
Moisture (%)	8.74	9.77	8.54
Crude protein (%)	26.27	30.04	27.66
Crude fiber (%)	15.97	19.88	12.42
Ether extract (%)	11.26	9.45	9.50

Water quality parameter

The temperature varied between 18.2°C and 31.8°C, with an average of 26.02±1.57°C during the morning hours (8-9 am). In the afternoon (3-4 pm), temperatures ranged from 21.2°C to 34.1°C, with an average of 28.86±1.44°C (Table 3). Dissolved Oxygen (DO) level ranged from 3.2 mg/L to 8.9 mg/L, averaging 5.13±0.22 mg/L in the morning, while in the afternoon, it ranged from 7.5 mg/L to 8.9 mg/L, with an average of 7.74±0.20 mg/L. The pH levels ranged from 7.3 to 8.5, with an average of 7.8 in the early morning. In the afternoon, the pH ranged from 8.5 to 8.8, with an average of 8.3.

The temperature, DO, and pH values were within the acceptable ranges for tilapia culture. The average DO values were higher than the minimum DO requirement of 4.0 mg/L for tilapia, indicating an adequate oxygen supply in the water. The pH values were also within the optimal range of 7.0-9.0, suitable for tilapia cultures (Table 4). Monitoring of water quality parameters ensured similar conditions in the cemented tank and minimized their impact on the growth and survival of fish.

Table 4. Range of some important water quality parameters recorded at the morning and afternoon during the experiment

Parameter	Unit	Time	Average	Range

Temperature	°C	8-9 am	26.02 ± 1.57	18.2-31.8
		3-4 pm	28.86 ± 1.44	21.2-34.1
DO	mg/L	8-9 am	5.13 ± 0.22	3.2-8.9
		3-4 pm	7.74 ± 0.20	7.5-8.9
pH	-	8-9 am	7.8	7.3-8.5
	-	3-4 pm	8.3	7.5-8.8

Fish growth performance, survival, and other parameters

The overall growth and survival parameters of Nile tilapia is shown in Table 5. In the present experiment, growth trends of *O. niloticus* fingerlings across all treatment groups tend to be positive. Among the different diets tested, the fingerlings that were fed T2 exhibited the most impressive growth performance (Fig. 1). This was reflected in their average weight gain (AWG), daily weight gain (DWG), all of which were significantly higher compared to those of the other diets. The results obtained showed that the average final weight (g/fish) of fish in T2 was significantly higher ($p<0.05$) than that of T1 and showed no significant difference ($p>0.05$) with T3. Similarly, the average total weight gain (g/m^2) of fish was significantly higher compared to T1 and had no significant difference ($p>0.05$) with other treatments. Similarly, the DWG of T2 was significantly higher ($p<0.05$) than that of T1, while no significant difference with T3. The apparent feed conversion ratio(AFCR) The apparent feed conversion ratio (AFCR) was lowest in T3 (1.94 ± 0.02), indicating better feed utilization efficiency. In contrast, the feed conversion efficiency (FCE) was lowest in T1 (0.48 ± 0.012), meaning only 48% of the feed was converted into fish biomass compared to other diets. The survival rate of the stocked fish seed ranged from 97.8 to 100%. The survival rate of T3 was higher (100%) than the other two diets.

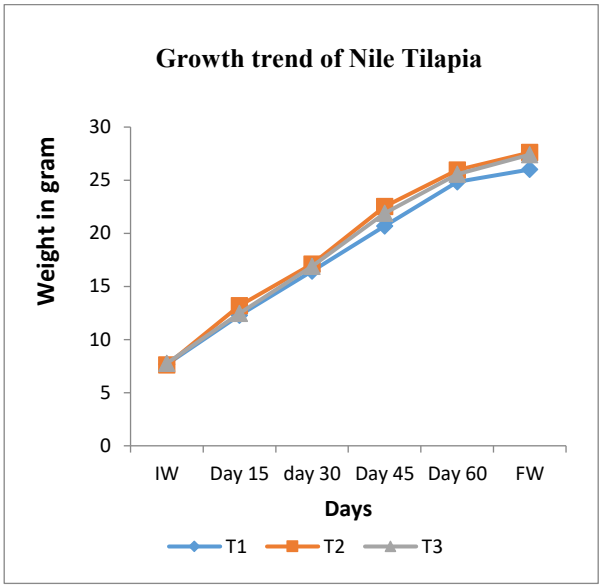

Figure 1. Growth trend of *Tilapia niloticus* fed with three different diets in the present experiment

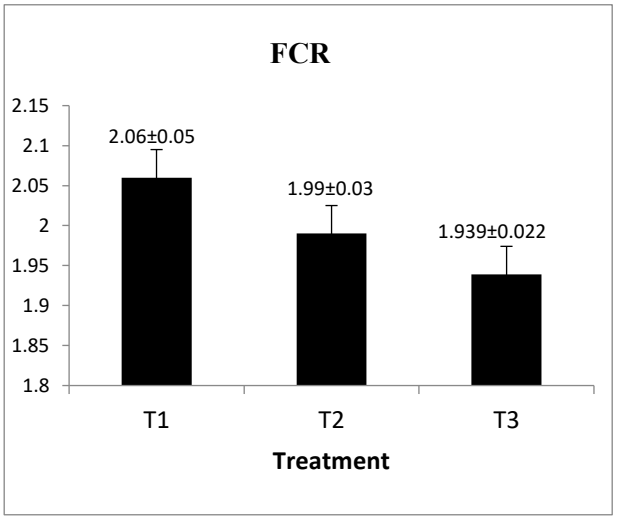

Figure 2. FCR of Nile Tilapia

Table 5. The growth performance of *Oreochromis niloticus* (Nile tilapia) fed with three different plant-based diets.

Treatment	Diet A	Diet B	Diet C
IW	7.66±0.088	7.63±0.18	7.76±0.09
FW	26.01±0.10^a	27.60±0.10^b	27.40±0.06^b
AWG	18.33±0.17^a	19.96±0.27^b	19.63±0.13^b

DWG	0.245±0.003[a]	0.266±0.004[b]	0.262±0.002[b]
Feed intake (g)	1095.333±12.732[a]	1173±19.399[b]	1142.33±5.544[ab]
FCR	2.06±0.051	1.99±0.034	1.94±0.022
FCE	0.486±0.012	0.504±0.009	0.516±0.006
SR (%)	97.76±2.233	98.90±1.10	100±0.00

Note: Values are presented as the mean ± SE. Values within the same row with different letters are significantly different (P<0.05).

Discussion

The findings of this study indicate that *Oreochromis niloticus* fingerlings readily accepted all three plant-based experimental diets. This suggests that the processing methods employed may have neutralized potential anti-nutritional factors, resulting in good feed palatability and nutrient utilization. The consistent acceptance and growth performance across treatments support the potential of plant-based proteins in tilapia diets. Although this is a preliminary finding, a long-term comprehensive study might be desirable to rule out this assumption.

Among the tested diets, the highest weight gain was observed in fish fed the soybean-enriched diet (T2), followed by the moringa-enriched diet (T3). Although differences in Specific Growth Rate (SGR) were not statistically significant ($p > 0.05$), fish on T2 achieved the greatest final weight, daily weight gain (DWG), and absolute weight gain (AWG). This aligns with previous research supporting the high protein digestibility and growth-promoting properties of soybean meal (Tacon et al., 2011; Reigh & Ellis, 1992; Mothwa et al., 2013).

Feed Conversion Ratio (FCR) was lowest in the moringa-enriched group (T3: 1.94 ± 0.022), indicating better feed utilization, although no significant differences were noted across treatments. These results are in line with earlier studies demonstrating the potential of *Moringa oleifera* leaf meal as a partial fish meal substitute in *O. niloticus* diets (Afuang et al., 2003; Parveen et al., 2021). While T2 promoted superior growth, T3 offered efficient feed conversion, suggesting moringa's utility in supporting feed efficiency and survival.

Survival rates remained high and statistically similar across all treatments, with T3 achieving 100% survival. This may indicate a health-supporting effect of *M. oleifera*, known for its antioxidant and immune-boosting properties. The comparable SGR across diets is likely due to the similar crude protein content (26–30%), which falls within the optimal range for tilapia growth (Balarin & Haller, 1982).

Water quality parameters remained within optimal ranges for tilapia culture throughout the study (DO: 3.2–8.9 mg/l; pH: 7.3–8.8), minimizing potential environmental influence on growth and survival (El-Sherif & El-Feky, 2009; Bahnasawy et al., 2003).

The inclusion of rice bran and mustard oil cake as base feed components, supplemented by soybean or moringa, provided a balanced nutrient profile. While fish can absorb some minerals from water, dietary supplementation remains essential for consistent growth and development (Craig et al., 2017). This study was of short duration and conducted under controlled tank conditions. Long-term trials across diverse environments, including commercial-scale operations, are needed to validate these findings. Further research should also evaluate cost-effectiveness, gut health, and immune responses linked to moringa-based diets.

Conclusion

Overall, soybean-enriched diets supported the highest growth performance, while moringa-enriched diets offered superior feed conversion and 100% survival. Both plant-based feeds demonstrated potential as sustainable alternatives to fish meal, with soybean being

more suitable for maximizing growth, and moringa contributing to efficient feed use and fish health. These findings support the integration of locally available plant proteins into sustainable aquaculture practices in Nepal

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