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Effect of different levels of *Moringa oleifera* on growth and nutritional performance of Rohu, *Labeo rohita* (Ham.1822)

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Abstract
The present study on the effect of different levels of *Moringa oleifera* leaf meal on growth and nutritional performance of rohu fingerling, *L. rohita* (average weight 7.12±0.75g) was evaluated. *Moringa oleifera* leaves were used at 0.00% (control) 4%, 8%, 12%, 16% and 20% levels by replacement of basal feed in treatment diets: T_0, T_1, T_2, T_3, T_4 and T_5 respectively. Feeding trial was conducted in FRP tanks of 200-liter capacity with each treatment having three replications. Fish fed with the diets T_3, recorded significantly ($P \leq 0.05$) best growth performance, net weight gain and specific growth rate (SGR). Significantly reduced growth and nutritional performance were recorded in diets with elevated levels of moringa leaf meal in treatments T_4 and T_5. Feed–related mortality was not recorded during the feeding trial. The study revealed that *Moringa oleifera* leaf meal may be included in the diets of *L. rohita* fingerlings at level of 12% by replacement of equal quantity of basal feed. Therefore, reducing the cost of fish meal in aquafeed. It is recommended that *Moringa oleifera* leaf meal may be used to partially replace the expensive source of protein i.e. fish meal, ground nut oil cake and soybean meal in the diets of *L. rohita* fingerlings.

Keywords: feed, mortality, specific growth rate, moringa, rohu

Introduction
The aquaculture industry has been globally recognized as the fastest growing food producing industry. Aquaculture production has continued to show strong growth, increasing at an average annual growth rate of 5.8 percent, per year during the period 2000-2016 [8]. This increase in aquaculture production further brings about higher growth in the number of aqua culturists, which was more than 10 million people in 2005 to 20.5 million in 2018 [9]. However, the major challenge with the growth recorded in aquaculture is the over dependence of aqua feeds on fish meal. Fish meal is one of the most expensive ingredients of aquaculture diets. Aqua culturists have therefore, begun to evaluate alternative diet ingredients to replace costlier protein source with readily available inexpensive plant sources. Considerable emphasis has been focused on the use of conventional plant protein sources, such as soybean [16, 20], groundnut [16], cottonseed [16, 6] and rapeseed meal [16] to replace fish meal in the diet of fish. However, their scarcity and competition from other sectors for such conventional crops for livestock and human consumption as well as industrial use make their costs too high and put them far beyond the reach of fish farmers or producers of aquafeeds [10]. The *Moringa oleifera* tree is widely distributed in the tropics. It holds a considerable potential for becoming an ingredient for animal and fish because of its high nutritional quality that is comparable to other feed protein source [4, 1]. *Labeo rohita* is one of the most important major carp species cultured in India on large scale. It is grown under poly-culture system with the other species of major and chinese carps [14]. Regional culture practices are mostly based on semi-intensive culture systems. It is the common inhabitant of the riverine system of the northern and central India. Information on its culture is available only from the early part of the 20th century. Its high growth potential with excessive consumer preferences, have accepted rohu as the most important cultivable freshwater species in India. There is scarcity of information regarding the utilization of Moringa leaves in the diet of carps. Therefore, the aim of this study is to determine the effects of dietary *Moringa oleifera* supplementation on the growth and nutritional performances of rohu (*L. rohita*) fingerlings.

Materials and Methods

The present study was carried out in the department of aquaculture, college of fisheries, Maharana Pratap university of agriculture and technology, Udaipur, Rajasthan. The experiment consisted of six treatments with each representing different inclusion level of *Moringa oleifera*. Those treatments were with 3 replications. The control had no inclusion of *Moringa oleifera*. The different level of *Moringa oleifera leaf* meal used were 0.00% (control) 4%, 8%, 12%, 16% and 20% by replacement of equal quantity of basal diet in treatments T_0, T_1, T_2, T_3, T_4 and T_5 respectively.

Experimental fish

Labeo rohita fingerlings (average weight of 7.12±0.75g) were obtained from the hatchery of aquaculture research unit, MPUAT. The fishes were first conditioned to the experimental water and temperature in the laboratory for a period of 15 day maintained on basal diet. The fingerlings were not fed for 24 hours before they were released in the experimental tanks to maintain a uniform stomach condition of fish and to induce their appetite for the commencement of the feeding trial [10]. After initial weighting the fishes were weighed fortnightly and the average weight was recorded to decide the daily ration.

Preparation of experimental diet

The basal diet was prepared by mixing different ingredient *viz*. Ground nut oil cake, Soybean meal, Fish meal, Rice bran, Veg. oil (Table 1). Leaves of the *Moringa oleifera* were collected from Horticulture farm, RCA, MPUAT, Udaipur and identified by the botanical expert of horticulture department of RCA, Udaipur. After collection, the leaves were cut into small pieces, soaked in water to remove dust and sun dried. Different ingredients used to prepare fish diet were analyzed for crude protein, fat, moisture, ash, crude fiber using (Table 1). *Moringa oleifera* leaf meal was measured out and mixed with basal feed at a ratio of 0.00%, 4% 8%, 12%, 16% and 20% as explained in Table 2. The fishes were fed at the rate of 4% of their body weight per day at 9-10 AM.

Table 1: The details of the ingredients used for basal diet (g/100g).

S. No.	Ingredients	Crude protein %	Basal diet %	
			3	Crude protein (g)
1	Ground Nut Oil Cake	40	18	7.2
2	Soybean meal	50	15	7.5
3	Fish meal*	55	20	11
4	Moringa Leaf Meal	22	0	0
5	Rice Bran	14	35	4.9
6	Vegetable oil	0	07	0
7	Tapioca Powder	0	03	0
8	Vitamin/Mineral	0	01	0
9	Chromium oxide	0	01	0
10	Total	181	100	30.6

Table 2: The details of experimental diets used for the experiment (g/100gm)

S. No.	Diets	Basal diet (g)	Moringa leaf Meal (g)	Total(g)
1	Control T_0	100	-	100
2	Treatment T_1	96	4	100
3	Treatment T_2	92	8	100
4	Treatment T_3	88	12	100
5	Treatment T_4	84	16	100
6	Treatment T_5	80	20	100

Analytical procedure

The analysis for proximate composition of diets used in the experiment were performed according to the procedure of the AOAC (2010) [3].

Evaluation of growth performance

Analysis of fish growth performances during the experiment were based on productivity indices on growth performance as described by Fasakin (1997) [10] and were as follows:

Total feed intake

This was estimated by adding up the fortnightly feed intakes during the experimental period.

Total weight gain

This represents the difference between the initial weight and the final weight gained.

Total weight gain = final weight – initial weight

Total percent weight gain (%)

This was calculated using the formula

TPWG = Total weight gained / Initial weight x 100%

Specific growth rate (SGR)

Specific Growth Rate was calculated from the relationship of the difference in the weight grain of fish within an experimental period.

Results

Proximate composition of feed

The proximate composition of the experimental feed was for the six treatments with different levels of *Moringa oleifera* leaf meal T_0 (0)%, T_1 (4%), T_2 (8%), T_3 (12%), T_4 (16%) and T_5 (20%). The proximate composition of the experimental diets is presented in Table 3.

Table 3: Proximate composition (in percent) of feed for *L. rohita* fingerlings in different treatments

Treatments	Protein	Fat	Carbohydrate	Ash	Moisture
T_0	29.54	9.42	48.72	5.50	6.82
T_1	29.30	9.40	48.97	5.54	6.79
T_2	29.01	9.36	49.32	5.56	6.75
T_3	28.75	9.30	49.41	5.64	6.90
T_4	28.49	9.28	49.63	5.68	6.92
T_5	28.23	9.24	49.57	6.00	6.96
SEm±	0.15	0.02	0.19	0.07	0.04
CD at 5%	0.46	0.06	0.59	0.22	0.14
CV (%)	0.89	0.36	0.67	2.18	1.12

There was an upward trend in the moisture and ash content of fish with increasing moringa inclusion at the end of the study. However, crude protein content was ranged from 28.23-29.54% in all experimental groups. Although slight decline in protein content was recorded with gradual increase in moringa leaf meal in diet.

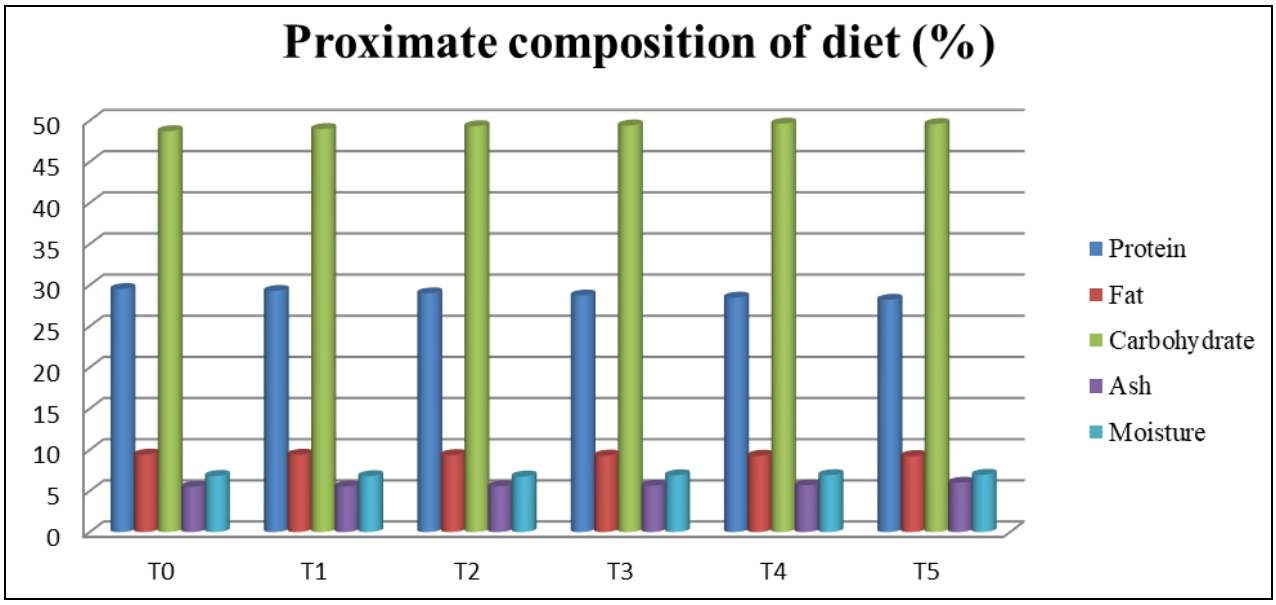

Fig 1: Proximate composition of experimental diet in percent

Growth performance and nutrient utilization of *L. rohita* fingerlings fed *Moringa oleifera* diet

The result of growth performance and nutrient utilization of *L. rohita* fingerlings fed varying levels of *Moringa oleifera* meal are presented in Table 4. In the feeding trials, growth parameters like weight gain, percentage weight gain and specific growth rate in treatment T_0, T_1, T_2, T_4 and T_5 were significantly lower than the T_3 ($P<0.05$). The fishes in the treatment T_3, fed better during the entire period of the experiment than in other treatments lowing to higher weight gain. Mortality was not recorded in any treatment throughout the 60-day feeding trial.

Table 4: Cumulative growth performance of *L. rohita* fed varying *M. oleifera* leaf meal inclusion levels

Parameter	Final Weight	Initial Weight g)	Weight Gain	Weight Gain %	Feed Fed	SGR	*Mortality %*
T_0	14.15	7.10	7.05	99.266	27.90	1.149	0
T_1	14.43	7.01	7.42	105.960	28.12	1.204	0
T_2	14.85	7.12	7.73	108.623	29.03	1.226	0
T_3	15.54	7.06	8.47	119.989	30.07	1.314	0
T_4	14.69	6.99	7.70	110.054	28.72	1.237	0
T_5	14.39	7.03	7.35	104.557	28.18	1.193	0
SEm±	0.03	0.03	0.04	0.907	0.04	0.0072	0
CD at 5 %	0.08	NS	0.11	2.796	0.13	0.0221	0
CV %	0.31	0.71	0.84	1.45	0.25	1.02	0

Discussion

The study has confirmed that the *M. oleifera* leaves have the potential to partly replace basal diet by and reduced expenditure on fish meal and other protein sources GOC, Soybean meal, without reducing growth and nutritional performances of the Rohu fingerlings. In view of the favorable amino acid profile of Moringa leaves and their wide and easy availability throughout the tropics it can be considered as a possible feed component with high nutritive value for fish [4]. Likewise, Abo-State *et al* ., (2014) [1] while evaluating the effect of feeding of raw moringa (*Moringa oleifera* Lam.) leaves meal (0%, 8%, 10% and 12%) to Nile tilapia (*Oreochromis niloticus*) fingerlings diets, found that it can be fed up to 8% level of dietary protein in without any negative effect on growth performance, nutrient utilization and carcass composition. It was concluded that moringa is one

of promising plant protein sources for aquaculture. In control to this the present study indicated that up to 12% replacement of basal diet with moringa leaf meal, it is beneficial for fish growth. Richter, *et al* ., (2003) [19] revealed that moringa leaf meal in the diets of *Oreochromis niloticus* at 10% inclusion did not affect growth, however, at high levels (20% or more), the growth of *O. niloticus* was negatively affected. In the current study, replacement of basal diet with moringa leaf meal at level as high as 16% (T_4) and 20% (T_5) reduced the growth rate and weight gain in *L. rohita* compared to 12% T_4 levels. As there was no mortality with moringa inclusion in all diets does not indicated that any adverse effect on the fish growth.

It has been reported that saponins in lupin seed meal and alfalfa impaired the growth performance of rainbow trout [13] and tilapia [18, 21]. A relatively higher concentration of total phenolics from mucuna beans (< 0.72%) in common carp diet has also been shown to significantly reduce the growth performance and feed utilization in common carp [2]. Protein digestibility and amino acid availability can also be reduced by the existence of antinutrients in fish feed [10]. Reason for low growth for end at high moringa inclusion levels might be combination of the antinutrients with fiber causing significant reduction in growth and nutritional performance of fish in treatment T_4 and T_5. The processing technique in this study which include soaking and drying might have reduced the soluble antinutrient in the moringa leaf meal, therefore increasing acceptability this agreed with the work of Madalla (2008) [17] and Fagbenro, *et al* ., (1999) [7] who found that different processing techniques resulted in better palatability and growth in fish. For instance, Ritcher, *et al* ., (2003) [19] stated that freeze-dried moringa leaf meal could replace 10% fish meal in the diet of *Oreochromis niloticus* without any deleterious effects on the growth of the fish, while Afuang, *et al*., (2003) [2] reported that solvent –extracted moringa leaf meal replaced 30% fishmeal in the same fish. These works showed that basal diet could be replaced at low levels i.e. 8-12% by moringa leaf meal protein without causing any adverse effect on fish growth. Water quality of experiment was ambient so no mortality was reported. Same trends were recorded by earlier researchers *viz*. J., Kavindra *et al* . 2019 [15], Gatiyala, *et al*., 2020 [11].

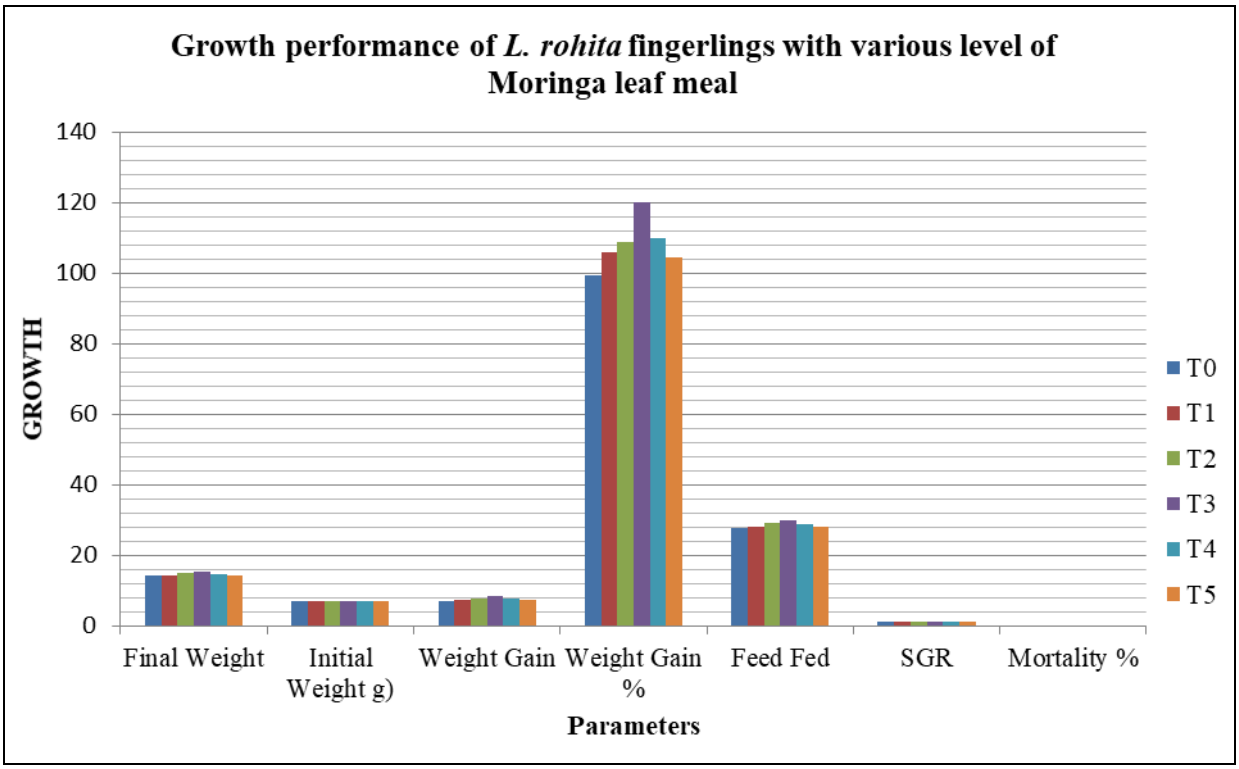

Fig 2: Growth performance of *L. rohita* fingerlings with various level of Moringa leaf meal

Conclusion

This current study identified moringa leaf meal as a non-conventional feed ingredient possessing many attractive attributes backing the results of this study, where 12% replacement of diet containg fish meal and soybean meal with moringa leaf meal supported good growth and nutritional performance of *L. rohita* fingerlings. However, more work may be conducted in finding methods for inactivating the antinutritional factors for inclusion of moringa leaves in practical diet for rohu fingerlings at higher levels.

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