

Fish growth performance, body composition and water quality in integrated system producing Grass carp (*Ctenopharyngodon idella*) in the Eastern Desert

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Abstract: The present work was conducted to evaluate the influence of replacing fish meal with agriculture residues on the water quality parameters, biochemical composition, and growth performance of grass carp (*Ctenopharyngodon idella*). A total of 400 grass carp (*Ctenopharyngodon idella*) with an average body weight of 100.1±1 g were used in the present study. The fish were randomly divided into 4 groups (100 fish/pond). A basal control diet was formulated to fulfil the nutrient requirements of the fish that contained 25% crude protein (CP). The other 3 diets (treatment diets) were alfalfa diet, peanut leaves diet, and a mixture of alfalfa and peanut leaves diet. The fish were fed six days per week for 90 days at a daily feeding of 3% of the estimated fish-weight of the total biomass of the fish with twice daily feeding at 8.00 am and 4.00 pm. Water quality parameters were found within a suitable range for fish production in the present experiment. Crude protein and moisture contents were the best when fish fed Alfalfa. Ash content showed significant differences among the tested diets. In addition, the lowest retention of crude protein and moisture at fish fed Peanut leaves.

The obtained data showed that growth performance was significantly affected by using alternative diet (Alfalfa, Peanut leaves and Alfalfa + Peanut leaves). As well as fish reared on Alfalfa + Peanut leaves showed the highest final weight (225.3 g), weight gain (91.6 g). While fish reared on Alfalfa showed lowest final weight (202.6 g), weight gain (79.3 g). It is observed that the best of FCR was recorded in fish fed on Alfalfa + Peanut leaves (1.7) followed by fish fed on Peanut leaves (1.9), whilst the worst FCR (2.1) was obtained in fish fed Alfalfa compared to the control group (1.3). Fish reared fed commercial diet exhibited the lowest significant survival percent (88.2%). Significant elevation was observed in the survival rate of the fish fed on alternative diet (Peanut leaves 88.6, Alfalfa + Peanut leaves 83.6 and Alfalfa 99.1) compared to the control. The result from this study indicates that alfalfa and groundnut leaves can be used as fish food in the diet of grass carp without adversely affecting growth efficiency and whole body composition.

Keywords: Grass carp, growth performance, body composition, water quality, aquaculture, Eastern Desert .

1 Introduction

Many researchers in Egypt's feed industry have been lured to the utilization of unusual, lower-cost agro-industrial by-products in order to produce high-quality commodities that fulfill customer demands while being cost-effective [1]. Animal and vegetable protein sources have been studied as alternative feedstuffs in fish feed production with varying degrees of effectiveness [2,3,4]. Because of their increased natural availability, researchers are particularly interested in vegetable products as components for fish feed production [5, 6]. Use of plant protein in fish feed industry has been tried for various commercial culture fish species as the formulation of feed is specific to species based on their specific requirements. The advantages lie not only in the availability and economic benefits but also that these plant

products also have less amount of phosphate and nitrogen than animal protein, therefore, reducing the chances of eutrophication of pond. Studies on green plant leaves as dietary sources for fish have focused on the use of leaf protein concentrates such as rye grass and alfalfa leaf protein concentrate [7, 8]. Among plant-derived raw materials, legumes have received special attention due to their high protein content. This has led to their use as fish meal replacements in tilapia feeds at levels as high as 100% in the case of soybean meal, or at lower levels when using novel sources such as *Leucaena leucocephala* leaf meal [9,10], alfalfa meal (*Medicago sativa*) [11,12] and protein concentrates [8,13,14]. *Medicago sativa* or alfalfa is a flowering plant grown throughout the world mostly as forage for cattle due to its high protein content and balanced amino acid profile, vitamins, and carotenoids [8,15]. Both

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fresh and dried leaves have experimented in the feed of tilapia [8]. In addition, Because of their high protein and energy content, peanuts or groundnuts (*Arachishypogaea*) are vital in human nutrition. In the 20 countries that produce the most peanuts in the shell, annual production exceeds 33 million tonnes [16]. Peanut plants yield high-protein fodder that has historically been utilized as pig and bovine feed, and its lowers are used as a nectar source in apiculture, in addition to the seed. Peanut stem and leaf production can reach 6 tonnes of dry matter per hectare (without seed production) [13]. From a nutritional perspective such protein concentrates have proven beneficial for only a limited number of cultured fish species. However, their potential usefulness may still depend upon the cost of their extraction and preparation [8].

The optimum water quality environment is important for the success of a commercial aquaculture system providing the rapid growth at the minimum cost of resources. It is important also in intensive recirculation systems to manage the aquatic environment and to optimize fish health and growth rates. These parameters include temperature, pH, dissolved oxygen, ammonia, nitrite, CO₂, and alkalinity. Each parameter is important, but it is the aggregate and interrelationship of all the parameters that influence the health and growth rate of the fish [17]. The relationship between water quality factors and their effect on fish growth rate and health is complicated. Environmental temperature changes affect the fishes' rate of biochemical reactions, which leads to different metabolic and oxygen consumption rates [18].

To identify economical and locally available feedstuffs, this study was designed to evaluate the use of crude plant leaves in formulated diets for grass carp. The plants used were alfalfa and Peanut leaves. Hence, the objective of the present study was designed to examine the effect of residues as feed for Grass carp (*Ctenopharyngodon idella*) in an integrated agri-aquaculture system in the Eastern Desert and its Influences on growth performance of Grass carp.

2 Materials and methods

Experimental Design

The present study was carried out on a private agriculture farm in the desert of Qena valley, Qena Governorate, Egypt Fig. (1). It is supported by water pond (reservoir) to secure the water needs for plants. This pond (2*2*1 meters) is lined, provided by water from an underground water well that is supplied by machine. About a sum of 400 fingerlings of fish grass carp (*Ctenopharyngodon idella*) was bought from Alahaywh governmental hatchery (General Authority for Fish Resources Development) in Sohag and transported under suitable conditions at early morning to secure its transportation during the long distance with the consideration of 10-20% loss. In the farm, the fish

fingerlings have been checked again and acclimated before being released to fishpond water. The fish were divided into 4 groups. The control group were fed on the commercial extruded floating fish diet (25% crude protein), the second group fed on alfalfa, the third group fed on peanut leaves the fourth group fed on a mixture of alfalfa and peanut leaves a six days per week for 90 days at a daily feeding of 3% of the estimated fish-weight of the total biomass of the fish according to [19] with twice daily feeding at 8.00 am and 4.00 pm Table (1). Feed quantity was adjusted according to the average body weight of the sample. The amount of feed was changed to the new fish biomass.

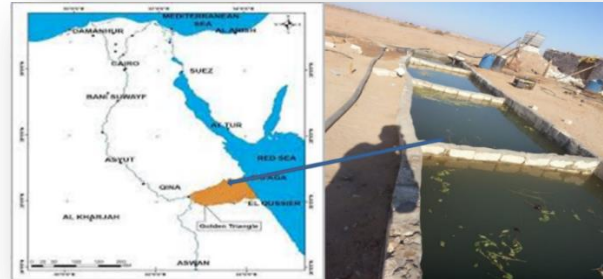


Fig. 1: a private agriculture farm in the desert of Qena valley, Qena Governorate, Egypt.

Water quality

For applying the integrated aqua-agriculture system in this farm, the water quality of the pond was analyzed to check its suitability for fish farming. The water samples were taken to analyze some chemical parameters according to standard method for examination of water and waste water, American Public Health Association [20] and according to the decisions of Ministry of Health and Ministry of Environment (No 92 - low 66 year 2013). These parameters included ammonia, dissolved oxygen, total hardness, salinity, phosphate, sodium, chloride, potassium, alkalinity, fluorine, sulphate, iron, pH values and temperature.

Growth performance parameters

For determining the trend of growth performance after the feeding trial, all growth parameters (final weight (FW), Survival rate (SR), weight gain (WG), specific growth rate (SGR) and feed conversion ratio (FCR) was measured monthly per individual using the following equations:

$$\text{Survival rate} = \frac{\text{Number of live fishes} \times 100}{\text{Total initial number of fish}}$$

$$\text{Weight Gain WG (g)} = \text{final fish weight (g)} - \text{initial fish weight (g)}$$

$$\text{Specific growth rate (SGR \%)} = \frac{\log \text{FW} - \log \text{IW}}{t} \times 100$$

Where FW is the final weight of fish (g), IW is the initial weight of fish (g) and t = total number of experimental days.

Feed conversion ratio (FCR) = feed intake (g) / weight gain (g).

Biochemical composition

Proximate composition analyses of diets and fish including moisture, protein, lipid, and ash contents were performed according to standard AOAC (2000) methodology on dry were CF, NFE weight basis as follow:

Moisture content

Water content was determined by drying a pre-weighed sample into an oven, thermostatically regulated, at 105°C overnight and then weighed until constant weight (complete dryness). The difference between the final and initial weights represented the water content of the sample.

% Moisture = 100 [weight loss (g)/weight of sample (g)]

Crude protein content (CP)

Protein content (on dry weight basis) was estimated as the total nitrogen content using the semi-automatic kjeldahl (Model VELP Scientifica, UDK 127). The Kjeldahl method divided into three main steps including digestion, distillation, and titration. Digestion is accomplished by boiling a homogeneous sample in 5 ml concentrated sulfuric acid (95%), the end result is an ammonium sulfate solution. Distillation is accomplished by adding excess base (NaOH) to the digestion product to convert NH_4 to NH_3 . Titration quantifies the amount of ammonia in the receiving solution. The amount of nitrogen in a sample can be calculated from the quantified amount of ammonia ion in the receiving solution. The ammonia is captured by a carefully measured excess of a standardized acid solution (H_2SO_4) in the receiving flask. The excess of acid in the receiving solution keeps the pH low, and the indicator does not change until the solution is "back titrated" with base (NaOH). Protein content was then calculated by multiplying total nitrogen by 6.25.

% protein = % Nitrogen content * 6.25.

Total lipid content

Lipid content was determined on wet weight basis using a Soxhlet apparatus with petroleum ether as the extraction solvent. Replicate 2 g of the lyophilized samples were wrapped in ashless N-free filter paper and extracted with petroleum ether in a side arm Soxhlet to a constant weight (16 hr) for ether extraction of lipid followed by drying in a muffle furnace at 60°C for 6-12 hr. Crude fat was calculated as the difference between lyophilized and extracted sample weight.

% Lipids = 100 [weight of lipid (g)/ weight of sample (g)]

Ash content

Homogenized diet or tissue samples were weighed into acid-washed glass ignition test tubes, 100 μL of 0.36% magnesium nitrate was added as ashing aid. The samples were then dried at 130°C for 2 hr and then ashed overnight in a muffle furnace with a dry ashing temperature of 480°C. When the sample ash was completely white, samples were diluted to 10 mL with 1 mol/L nitric acid and analyzed with a Perkin- Elmer 2380 Atomic Absorption Spectrophotometer equipped with both flame and graphite (PE HGA-400) furnaces. The difference between final and initial weights equals the ash content.

% Ash content = 100 [weight of ash (g)/ weight of dry sample (g)]

Statistical Analyses

Data were presented as mean \pm SD. The results were subjected to one-way analysis of variance (ANOVA) to test the effect of treatment inclusion on fish performance. Data were analyzed using SPSS program, Version 16. Differences between means were compared using Duncan multiple range test at $P < 0.05$ level.

3 Results

Water quality

The water quality parameters of the fishpond during the experimental period were evaluated in two phases. The first one was to measure the suitability of the well's water for fishpond; they were in suitable range to culture grass carp (*C. idella*). These parameters were as follow water temperature record 25.9 °C for well water compared to 26.3 °C for pond (1), 26.1 °C for pond (2), 26.2 °C for pond (3), 26 °C for pond (4); maximum pH value from ponds was 7.8 mg/l for pond (1) and decline to 7.58 mg/l for ponds (2) and (4), maximum dissolved oxygen (DO) 5.3 mg/L for pond (2) and the lowest was 4 mg/L for pond (1) compared to 4 mg/L for well water; Total Hardness 127 mg/l for well water to be the lowest value 119 mg/l for pond (4), and total dissolved salts TDS 1151 mg/l for well water and pond (2) to be 1148 mg/l for pond (3) (Table 1). The second phase was operated to monitor the changes in water quality that will be used for plants. The parameters of the four ponds are shown in (Table1). From the table, the ionized ammonia (NH_4) was recorded to be 6.2 mg/l for pond (1), 5.2 mg/l for pond (2), 5.7 mg/l for pond (3) and 5.9 mg/l for pond (4), the unionized ammonia (NH_3) was recorded to be 3.5 mg/l for pond (1), 3.5 mg/l for pond (2), 3.4 mg/l for pond (3) and 3.6 mg/l for pond (4), Phosphate (PO_4) 0.5 mg/l for pond (1), 0.6 mg/l for pond (2), 0.6 mg/l for pond (3) and 0.5 mg/l for pond (4), Phosphorus (P) 0.3 mg/l for pond (1), 0.1 mg/l for pond (2), 0.3 mg/l for pond (3) and 0.2 mg/l for pond (4), Iron (Fe) 0.2 mg/l for pond (1), 0.1 mg/l for pond (2), 0.1 mg/l for pond (3) and 0.1 mg/l for pond (4), Sodium

(Na) 160 mg/l for pond (1), 152 mg/l for pond (2), 157 mg/l for pond (3) and 152 mg/l for pond (4) and Potassium (K) 91 mg/l for pond (1), 88 mg/l for pond (2), 87 mg/l for pond (3) and 88 mg/l for pond (4). These parameters were higher than those measured in well's water. While the following parameters; Tot. MgCo₃ 47 mg/l for pond (1), 48 mg/l for pond (2), 47 mg/l for pond (3) and 47 mg/l for pond (4) and ALK 162 mg/l for pond (1), 160 mg/l for pond (2), 160 mg/l for pond (3) and 161 mg/l for pond (4) were lower than those measured for well water (Table 1).

Evaluation of growth performance.

The changes in mean body weight (g/fish) of grass carp fed on four tested diets including: basal diets, Alfalfa, Peanut leaves and Alfalfa + Peanut leaves during the period of the experiment (3 months) are shown in (Table 2). At the

end of the experiment, the mean weight ranged between 202.3 ± 13.7 g at the control group and 225.3 ± 2.5 g at Alfalfa + Peanut leaves group. The overall data of final weight, weight gain and specific growth rate (SGR) for grass carp reared at four diets for a period of 3 months are presented in (Table 2). It can be concluded from this Table 2, mean final weight (g/fish), weight gain (g/fish) and SGR (%/day) were significantly affected and the best mean final weight (g/fish), weight gain (g/fish) and SGR (%/day) were obtained with the Alfalfa + Peanut leaves feeding. While the result of feed conversion ratio is presented in table 3. Feed conversion ratio was significantly ($P \leq 0.05$) affected by plant feed and exhibited the best results at the Alfalfa feeding. The feed conversion ratio (FCR) of Nile tilapia fed on Peanut leaves and Alfalfa + Peanut leaves were not significantly different ($P > 0.05$) when compared with control diet.

Table (1):Parameters of water quality of the well water and fishponds water

| Parameter | Well water | Pond water (1) Control | Pond water (2) Alfalfa | Pond water (3) Peanut leaves | Pond water (4) Alfalfa + Peanut leaves | (MH and ME,2013) |
|-----------------------|------------|------------------------|------------------------|------------------------------|----------------------------------------|------------------|
| T (c) | 25.9 | 26.3 | 26.1 | 26.2 | 26 | |
| pH | 8 | 7.8 | 7.5 | 7.6 | 7.5 | 6.5-9mg/l |
| DO (mg/l.) | 4 | 4.0 | 5.3 | 4.9 | 5.1 | Min 4 mg/l |
| Hardness (mg/l) | 127 | 122 | 120 | 122 | 119 | 30-180 mg/l |
| F | 0.2 | 0.1 | 0.2 | 0.1 | 0.1 | 1mg/l |
| NH ₄ | NIL | 6.2 | 5.2 | 5.7 | 5.9 | NIL |
| NH ₃ | 0.9 | 3.5 | 3.5 | 3.4 | 3.6 | 2.4 mg/l |
| TDS | 1151 | 1150 | 1151 | 1148 | 1150 | 0-4000 mg/l |
| PO ₄ | NIL | 0.5 | 0.6 | 0.6 | 0.5 | 0.06 mg/l |
| BOD | 51 | 57 | 57 | 54 | 56 | 60 mg/l |
| P | NIL | 0.3 | 0.1 | 0.3 | 0.2 | NIL |
| NO ₃ | NIL | NIL | 0.1 | 0.1 | NIL | 45mg/l |
| Tot.CaCo ₃ | 81 | 83 | 81 | 86 | 83 | 63 - 250 mg/l |
| Tot.MgCo ₃ | 49 | 47 | 48 | 47 | 47 | 150 mg/l |
| Fe | 0.1 | 0.2 | 0.1 | 0.1 | 0.1 | 0.01 mg/l |
| Mn | 0.09 | 0.07 | 0.07 | 0.09 | 0.06 | 0-0.01 mg/l |
| CL | 439 | 438 | 435 | 439 | 438 | 500 mg/l |
| Na | 126 | 160 | 152 | 157 | 152 | 200 mg/l |
| K | 73 | 91 | 88 | 87 | 88 | 150 mg/l |
| ALK. | 170 | 162 | 160 | 160 | 161 | 50-300 mg L-1 . |

Table (2): Growth performance of fish of Grass carp *Ctenopharyngodon Idella* during the experiment.

| Items | Experimental diets | | | |
|--------------|--------------------|----------------------|--------------------|-------------------------|
| | Control | Alfalfa | Peanut leaves | Alfalfa + Peanut leaves |
| IW (g/fish) | 126 ± 8.7^a | 123.3 ± 9.0^{ab} | 123 ± 2.2^b | 133.6 ± 3.1^c |
| FW (g/fish) | 202.3 ± 13.7^a | 202.6 ± 8.2^a | 214 ± 10.5^b | 225.3 ± 2.5^b |
| TWG (g/fish) | 76.3 ± 13.1^a | 79.3 ± 2.6^b | 91 ± 10.6^{ab} | 91.6 ± 0.7^{bc} |
| SGR (%/d) | 1.6 ± 0.2^a | 1.9 ± 0.2^a | 2.1 ± 0.1^b | 2.4 ± 0.2^b |
| FCR | 1.3 ± 0.1^a | 2.1 ± 0.2^b | 1.9 ± 0.2^{ab} | 1.7 ± 0.2^{ab} |
| SR (%) | 88.2 ± 3.2^a | 99.1 ± 1.2^{ab} | 88.6 ± 1.2^a | 83.6 ± 1.5^b |

Biochemical composition of fish.

The effect of Alfalfa, Peanut leaves and Alfalfa + Peanut leaves on whole body composition of fish in the present experiment are illustrated in (Table 3).

The best moisture and ash contents were obtained with the Peanut leaves feeding (78.7 ± 20.9 , 3.4 ± 2.1 , respectively). While The best protein and crude lipids contents were obtained with the alfalfa feeding (14.5 ± 2.9 , 4.3 ± 2.5 , respectively).

Table (3):Body Composition of Grass carp (*Ctenopharyngodonidella*) in experimental treatments

| | Control | Alfalfa | Peanut leaves | Alfalfa + Peanut leaves |
|-----------------|-------------------|-------------------|----------------------|-------------------------|
| Moisture | 76.2 ± 22.3^a | 77.2 ± 15.3^b | 78.7 ± 20.9^{ab} | 78.4 ± 21.2^{ab} |
| Protein | 13.2 ± 3.3^a | 14.5 ± 2.9^b | 14.2 ± 3.2^a | 14.3 ± 4.6^b |
| Lipid | 6.9 ± 1.2^a | 4.3 ± 2.5^b | 4.7 ± 1.2^{ab} | 4.5 ± 3.2^{ab} |
| Ash | 4.7 ± 2.1^a | 4.5 ± 1.7^a | 3.4 ± 2.1^b | 3.9 ± 0.04^{ab} |

4 Discussion

The water quality of fish farm is among the most important factors affecting the success of fish farming, preventing mortality and improves the healthy status of fish. Most of the water quality problems can be solved with adequate water exchange. Thus, if large quantities of water suitable for aquaculture are available, monitoring would not be as critical and high production levels can be targeted. If water is limited, the risk of encountering water quality and disease problems increases as one goes for more intensive culture, [21]. The water exchange in fish farm is used to adjust salinity as desired, to remove excess metabolites, to keep algae healthy to produce ample oxygen and to regulate pond water temperature [22]. Water quality parameters were found within a suitable range for fish production in the present experiment. Temperature is an important water quality parameter which was found to be influenced by season. The suitable range of water temperature for freshwater aquaculture is about 25-32 °C [23]. The optimum water temperature recorded from 25.9 °C to 26.3 °C which was closer to [24] reported as 16.1 °C to 19.4 °C. [25] stated optimum water temperature of ponds ranged from 26.0 °C to 32.4 °C. The concentration of dissolved oxygen (DO) ranged from 4mg/l to 5.3 mg/l which was closer to those reported by [26] of 4.3 mg/l to 6.8 mg/l. [27] noticed the range of pH for pond water would be 7.5 to 8. The pH value in the experimental ponds varied from 7.5 to 8 which was closer to the finding of [28] ranged from 6.5-9 indicating the suitable condition for fish culture. [29] stated the maximum permissible limit of total dissolved solids for fish culture was 4000mg/l. The total dissolved solids in the present study were 1150 mg/l which was within desirable range for fish culture. Ammonia (NH₃) ranged from 3.4 mg/l to 3.6 mg/l during the experiment, which was more than 2.4 mg/l, based on [30] which stimulate plant growth. During grass carp feeding, excretion (consisting of approximately 80% ammonia, a little carbamide, and some indigestive plants) discharged by fish into water increased the concentrations of NH₄⁺ -N, NO₂⁻ -N, TN, and TP [31]. According to [32], plants in the integrated system act as biological filters, thereby absorbing nutrients such as nitrate and NH₃ from the system.

[33] and [34] suggested that the acceptable levels of nitrogen for plants at the outset are optimal at around, 100 mg/l, while it should be at 200 mg/l during growth. Well and spring water may contain elevated levels of iron (ferrous iron) and manganese, but still appear clear to the eye. Ordinarily, waters high in these elements can be used "as is" for outdoor culture ponds because the iron floc settles to the pond bottom and does not interfere with fish in the water column [35]. The iron concentrations in water source ranged from 0.1 to 0.2 mg/l. Nonetheless, the iron levels were only slightly above the recommended limit. [36] reported that iron concentrations less than 0.5mg/l would be appropriate for hatcheries and channel catfish and other warm water species while the optimal iron concentration for cold water temperature is less than 0.15mg/l. But [37] conservatively recommends a general standard of less than 0.01 mg/l.

Growth is a complex biological process affected by several factors such as physiological, behavioral, nutritional, genetic, and environmental conditions. The genetic factor (indigenous factor) of the target fish, providing sufficient amount of balanced diets, feeding rate and feeding frequency, stocking density and optimum environmental conditions (exogenous factors) are important to achieve maximum growth of the cultured fish [38]. In the present study, the obtained data showed that growth performance was significantly affected by using alternative diet (Alfalfa, Peanut leaves, and Alfalfa + Peanut leaves). As well as, fish reared on Alfalfa + Peanut leaves showed the highest final weight (225.3 g), weight gain (91.6 g). While fish reared on Alfalfa showed lowest final weight (202.6 g), weight gain (79.3 g). It is observed that the best of FCR was recorded in fish fed on Alfalfa + Peanut leaves (1.7) followed by fish fed on Peanut leaves (1.9), whilst the worst FCR (2.1) was obtained in fish fed Alfalfa compared to the control group. Diet affected moisture content of the yellow perch fillets in a small but measurable amount. Fish feed on the Alfalfa protein concentrate APC diet had 79.3% moisture content in the fillet as opposed to 78.9% moisture in fillets from fish on the fishmeal diet. Protein, lipid, ash, and energy content

of fillets did not change significantly with diet [39]. Alfalfa (*Medicago sativa*) is a flowering plant grown throughout the world mostly as forage for cattle due to its high protein content and balanced amino acid profile, vitamins, and carotenoids (8; 15). Both fresh and dried leaves have been experimented in the feed of tilapia (8). (8) reported that inclusion of alfalfa leaf protein up to 35% inclusion level can be used in feeds of tilapia without compromising the growth and survival of the fish. [40] reported that incorporating alfalfa protein concentrate in seabream feeds promotes growth. [41] stated that grass carp were fed with Alfalfa had positive effects on the growth parameters and had significant difference from other treatments. It has been reported enhanced growth and survival of common carp and mrigal coupled with an increase in protein and lipid content when alfalfa leaf meals are included up to 40%, 30% and 25 % respectively in the diet [42,43,15]. Also, fish-fed diets containing alfalfa leaf meals showed superior growth performance and body composition which was better than those fed control diet. The leaf meal-treated groups also had higher values of protein, lipid, energy retention and production of digestive enzymes amylase. Conclusively, the results indicated that leaf meal fiber provided better performance showing the inherent prebiotic effect of the utilization of these leaf meals in hybrid lemon fin barb [44]. [45] exhibited that grass carp fed on alfalfa had positive effects on the growth parameters compared with artificial diet. Most of the researchers recommended low replacement level. [12] suggested only up to 5% inclusion level. Adding of low levels of alfalfa in the meal enhances utilization of nutrient with growth performance in carp (*Cyprinus carpio*) fish and improving health performance as compared to the control by mean of biological parameters as the alfalfa leaves as feed additives for giving more time for fish to digest their feeds [46]. In other hand, [47] reported that complete replacement may hamper the growth performance. [11] indicated that the growth depression of fish fed diets supplemented with dehydrated alfalfa meal could be attributed to the presence of growth inhibitors such as a trypsin inhibitor which is reported to occur in vegetative tissues of several plants such as dehydrated alfalfa. The trypsin inhibitor activity impairs the digestion and absorption of protein [48;11]. These different results among studies may be explained by differences in digestion physiology between fish species and/or processing technology of the alfalfa. Since grass carp is an herbivorous and has a relatively long digestive tract which may allow for more efficient digestion of plant ingredients, alfalfa might be better utilized by this fish than the fish mentioned above.

The peanut is only one of a few hundred species of legumes that produces flowers above ground but develops the fruit below ground. The use of young peanut leaves as a green vegetable will increase utilization of the plant and reduce production wastes that will have to be recycled in an enclosed environment such as the controlled ecological life support system (CELSS) of a space station [49]. The usages of peanut products as a raw material for fish feed have not

been widely investigated. Lack of information exists about the usability of peanut meal instead of fish meal in aqua feeds. The level of acceptance or rejection of feed is a common problem when plant-based protein sources are used in aquafeed as an alternative feed stuff [50]. [51] stated incorporation of 10% or 20% peanut vines (PV) in *Clarias gariepinus* diet did not affect any parameter when compared with the control. [52] successfully replaced 20% of the fish meal diet with peanut leaf meal for Nile tilapia without negative effects on growth performance. [53] observed that low PNM inclusion level is possible (104-120 g kg⁻¹) in the diet for Pacific white shrimp. The study indicated that Groundnut (*Arachis hypogaea* L.) By-Products could replace at least 50% and 20% of fish meal protein, respectively, in the diet of *O. niloticus* fingerlings without adversely affecting growth, feed efficiency, and nutrient digestibility [54]. [55] indicated that the replacement of 20% dietary FM with PNM had no adverse effect on the growth performance, body composition parameters and general health of Mozambique tilapia fries. Also, [56] suggested that raw GNM could be used to replace up to 30% fish meal protein in Nile tilapia diets. The knowledge about the biochemical composition of fishes are plays vital roles in various fields [57]. Due to the unusual nutritional nature of fish the consummation of fish and alertness of balance diet food increases dramatically from day to day [58]. For the proper healthy and balance diet food it is essential to understand about the biochemical ingredient of fish which play the vital role for dieticians [59]. Animal used the fish and fish related products as source of food therefore it is extremely important for one to know the biochemical levels in fish body [60]. According to our results there were significant differences in moisture, protein, and ash content between the experimental treatments. [8] found that there was a slight reduction in body fat with increasing levels of chloroplastic alfalfa protein, which could have been due to dietary energy reduction; The same authors suggested that the nutritional quality of alfalfa LPC was adequate with respect for fish meal, particularly when cytoplasm protein was used. [12] stated that body moisture content of fish increased whereas fat and gross energy contents decreased when the level of alfalfa meal in diets was either 15 or 20%. No significant differences were observed in the body moisture, fat and gross energy contents of fish fed either 15 or 20% of alfalfa meal. [42] observed crude protein was increased in flesh of fish by the increase of alfalfa meal in the diet; while the opposite trend was observed for ether extract; ash % and gross energy (Kcal/kg) where they decrease as the substitution levels of alfalfa meal increased in the diets. The final whole-body composition of the experimental fish was broadly similar and relatively unaffected by the different dietary treatments with the exception of ash content, which was significantly lower for the diet with 50% Groundnut (*Arachis hypogaea* L.) By-Products [54]. [41] found there were not any significantly different in moisture, protein, and ash content between the experimental treatment. [55] stated the inclusion of peanut meal does not affect the proximate analyses of diets. The

lipid contents of the fish showed differences, but it is difficult to attribute this to the inclusion of PM in the diets. Addition of PM had no adverse effect on the composition of flesh protein and ash content. [56] observed that 30% inclusion level of Groundnut meal (GNM) are suitable replacements for fish meal in diets for fish.

In conclusion, the result from this study indicates that alfalfa and groundnut leaves can be used as fish food in the diet of grass carp without adversely affecting growth efficiency, and whole-body composition.

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