



Original Research Article

Nutrient Evaluation and Haematological Indices of Hybrid Catfish Fingerlings Fed Graded Levels of Germinated Sword Beans (*Canavalia gladiata*) Seed Meal

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This study was carried out to evaluate the growth response, nutrient utilization and body composition of hybrid catfish fingerlings fed graded levels of germinated sword bean seed meal for 56 days. 0% lipid with 0-45% inclusion of the GCGM at different graded levels to replace Clupeid fishmeal. The diets were designated as D1, D2, D3, D4 and D5. The concrete tanks were filled to 5/6 of their volume with filtered and dechlorinated tap water, and 20 fish with an initial average weight of 2.25 ± 0.02 g. Water temperature and other water quality parameters were monitored daily. The results at the end of the feeding trial showed that fish fed D4 had significantly greater growth parameter values: final body weight and percentage weight gain, while fish fed D5, had the lowest value in all the growth parameter indices measured. However, it was not significantly different from those of fish fed D1, D2 and D3. There was no significant difference in the percentage of survival among all fish fed the experimental diets. Fish fed D4 also had a higher and significant value in all the nutrient utilization parameter measures: The total feed intake, feed efficiency, protein efficiency ratio, and protein retention. Fish fed D5 had the lowest nutrient utilization value and was significantly lower than those of fish fed D1, D2 and D3. The proximate composition results revealed that carcass lipids increased with a proportional increase in the inclusion level of the GSBM meals in the diet. It could be concluded that 35% inclusion of GSBM meal improved growth performance and nutrient utilization of hybrid catfish without any adverse effects on their health status, suggesting that the GSBM meal could be a suitable ingredient in the diet of hybrid catfish.

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Article History

Received: 25th May 2021

Accepted: 12th July 2021

Published: 31st July 2021

Cite Article:

Bake, G. G., Gana, A. B., Nwangwu, D. C., Abdullahi, A., Igili, G. M. & Abdulkarim, I. A. (2021). Nutrient Evaluation and Haematological Indices of hybrid Catfish Fingerlings Fed Graded Levels of Germinated Sword Beans (*Canavalia gladiata*) Seed Meal. Journal of Research in Science and Technology, 2(6):54-65.

Keywords: Hybrid catfish, Growth performance, Sword bean, Seed meal, Hapas.

INTRODUCTION

Nigeria is one of the highest consumers of fish among the nations of Africa by far, owing to its mixture of large population and relatively high per capita consumption levels (Chan et al., 2019; Adeleke et al., 2020). The overexploitation of its natural water's resource is close to depletion, hence, little or no growth is expected from capture fisheries. Thus, fish production needs to be expanded to meet the demands of an ever-growing population (Oluwatobi et al., 2017). In Nigeria, fish production has gained much attention in comparison to other animal-producing sectors in terms of growth (Udo and Umanah, 2017). The procreation of African catfish (claridae family) is the lead of all aquacultural species produced in Nigeria. This species can be cultivated using small technological aquaculture system and they can eat wide range of feeds (Fagbenro et al., 2003). The claridae family and their hybrids possess high aquaculture values in the country these species are mainly cultured because of their high market value, fast growth rate and capacity to withstand harsh conditions especially low oxygen content and turbidity (Fagbenro et al., 2002). The reduction, inconsistency in supply, and sudden rising in the prices of fish meal and its co-product have triggered the search for viable alternatives for fish feed production.

Undoubtedly, fish feed is the most expensive input for aquaculture operations (Gatlin et al 2007; Enyidi et al., 2014). Feed takes about 60% – 70% of the entire production capital. The average farmers, especially in the developing nations are confronting with number of challenges for the need of high-class protein in fish food accompanied by soaring prices of fish meals and its co-products (Akinwande et al., 2002; Dada and Akinwande, 2005). Much of the high cost of feed arises from the extensive reliance on protein sources, such as FM and its co-products (Lim et al., 2004; Omoregie, 2008). The shortage and high cost of pelleted feed severely constrained the development of low-cost aquaculture systems suitable for small-scale farmers in developing countries. The decrease in the availability and increase in the prices of fish meals and its co-products have stimulated the probe for sustainable alternatives for aquaculture feeds (Olsen and Hasan, 2012). Given this, plant protein sources have continued to gain attention to satisfy the growing demands of the aqua-feed industry, as they can serve as a good alternative ingredient for partial or total replacement of fish meal in aqua-feed production. Among plant protein sources, the most conventionally used are legumes such as soybean, pea, lupin; corn gluten meal, and other cereal, concentrates (Gatlin et al., 2007). Furthermore, human reliance and competition from other sectors of the economy has also made these conventional feed ingredients unaffordable to an average fish farmer due to the sudden rise in their price as well as the ever-increasing demand for them. These have been seen as a threat to aquaculture sustainability in most developing countries especially Africa Nigeria

inclusive (Fasakin et al., 2003). To attain more economically sustainable and viable products as well as eco-friendly diets, research interest has been redirected towards the evaluation and use of unconventional under-utilized plants protein sources (Siddhuraju and Becker, 2001; Fasakin et al., 2003; Bake et al., 2009, 2015).

Although Plant Proteins (PP) may be relatively cheap, their use is usually limited by deficiencies in essential amino acids and minerals, and the presence of Anti-Nutritional Factors (ANFs), and a high level of non-starch carbohydrates (Francis et al., 2001; Fagbenro et al., 2004). The pursuit to use protein sources of plant origin as an alternate replacement for the fish meal component in fish diets has continued to show varying success. Ogunji and Wirth (2000, 2001) summarized some of the factors which may have led to the variations in the results obtained like; protein composition and amino acid profile of alternative feeds, nutrient utilization and digestibility of feeds, phosphorus content of alternative feeds, anti-nutritional composition of the substituted ingredients and the resultant diet as well as the palatability and acceptability of alternative feeds. Notwithstanding several industrial or home scale processes, such as soaking, germination, dehulling, milling, cooking, roasting or fermentation have been used to improve the nutritional properties of legumes. Although, the efficacy of these treatments have been found to varied (Alonso et al., 2000; Bake et al., 2015).

Improvement of plant-derived ingredients that will be very digestible/palatable and with less negative factors affecting digestion and metabolism as well as eco-friendly to the fish is of preeminent importance to fish feed researchers and producers. Most of these alternative ingredients can be utilized better when properly processed.

The germination processing method is an effective food processing method, an exclusive process that customarily boosts the nutritional quality of the feed ingredients and can decrease anti-nutritive factors, hence improves and increases the level and digestibility of free amino acids and available carbohydrates, enhancing mineral bioavailability, and improving the functional properties of cereal and pulses (Echendu et al., 2009). This food processing technique has the potential to provide a promising future for plant-derived ingredients for sustainable aquaculture (Bake et al., 2020). The culturing of Clariidae species has continued to generate great interest among fish farmers in Africa and Nigeria particularly and of recent trend is the culture of its hybrids with their aquaculture relevance. These could be attributed to their wide geographical spread, high growth rate, resistance to handling and other aquacultural practices and as well as been appreciated in a wide number of African countries (Eyo and Ezechie, 2004). The hybrids of *Heterobranchus* sp. and *Clarias* sp. exhibit the fast-growing quality of *Heterobranchus* sp. and resistant to diseases of *Clarias gariepinus* (Hogendoorn and Vismans, 1980; Ajana and Anyanwu, 1995;

Table 1. Proximate composition (on dry-matter basis) of the major ingredients used.

Ingredients	Fishmeal	Soybean meal	Maize meal	Millet meal	RCGM	GCGM
Proximate composition						
Moisture (%)	5.79	3.09	4.66	3.22	6.85	5.28
Crude protein (% d.b.*1)	65.34	43.63	9.32	12.9	28.61	35.82
Crude lipid (% d.b.*1)	11.36	7.00	4.20	4.36	9.44	8.83
Ash (% d.b.*1)	14.34	8.15	3.22	2.33	4.56	2.76
Crude fibre (% d.b.*1)	0.06	5.00	3.40	2.60	6.94	5.85

^aMeans of two replicate analyses.

Key: **RCGM** = Roasted *Canavalia gladiata* Seed Meal; **GCGM** = Germinated *Canavalia gladiata* Seed Meal.

Anibeze and Eze, 2000). One of the unconventional plant protein ingredients that may have the capacity for aquaculture use includes products from Sword beans (*Canavalia gladiata*) seed. *Canavalia gladiata* commonly called sword beans is a domesticated plant species in which the leaves resemble to swords. The seeds are taken as vegetable in Nigeria especially in the northern part of the country (amongst the Nupes, Bajus, Jabas, and Gbagis), it belongs to the Fabaceae family, a leguminous crop. However, the extent to which legume seed could be potential sources of protein and fish feed is limited due to the presence of anti-nutritional factors (ANFs). The presence of secondary metabolites such as phytin, tannin, Canavalia, con-canavalia-A, trypsin inhibitors to mention but a few, have been reported in raw sword beans (Akinmutimi, 2003). Although they possess a rich nutritional profile but little or no work has been done on processed (germinated) *Canavalia gladiata* seed meal which can be used as an alternative ingredient in the diet of hybrid catfish fingerlings. Thus, this study was conducted to evaluate the suitability of germinated *Canavalia gladiata* seed meal (GCGM) in the diet of hybrid catfish fingerlings through their growth performance, nutrient utilization, and haematological indices.

MATERIALS AND METHODS

Ingredients and diet formulation

Soybean Meal (SMB)

The main ingredients (Raw soybean) were bought from the Tunga market Minna (Niger state). The soybean was processed by toasting it in a frying pan at 80°C for 60 minutes until the color changes to golden brown and allowed to cool before milling with the aid of a grinding machine. Crude protein and lipid contents of SMB were 43.63% and 7.00%, respectively as shown in Table 1.

Fishmeal (FM)

The fishmeal used in this experiment was obtained from the Musgola Fish Farm, along the Bosso estate road Minna Niger State, Nigeria. The crude protein and lipid content of fishmeal were 65.34% and 11.36% respectively as shown in Table 1.

Germinated *Canavalia gladiata* seed meal (GCGM)

10 kg of raw sword beans (*Canavalia gladiata*) were collected from a local farmer Manchok Kaura local government of Kaduna. The raw beans were then sieved to remove dirt. 5kg of the seed was pre-soaked inside a bowl, the pre-soaking was carried out by mixing the *Canavalia gladiata* seed with water in the ratio of 3:1 (1 part of *C. gladiata* seed to 3 parts of water). This was done to separate the viable seed from the non-viable once, the non-viable seeds floated to the top surface of the water while the viable seeds remain at the bottom of the bowl. The viable seeds were then soaked inside warm water (80°C) overnight for 8 hours in a firmly sealed transparent plastic container. After 8hrs, the soaked seeds were removed and placed in a new bowl and covered with wet cotton wool at room temperature (29-30°C) for 72 hrs. The cotton wool was moistened with clean water at regular intervals of 12 hrs. The seeds that showed no sign of sprouting were discarded. The seed coats of the germinated (sprouted) seeds were removed and the seed cotyledons were washed and spread on a polythene sheet in a room and dried for six (6) days up to about 90% of the dry matter. The seeds were grinded into powder using a hammer mill. Crude protein and lipid contents of GCGM was 35.82% and 8.83%, while RSBM was 28.61% and 9.44% respectively as shown in Table 1.

All the ingredients were independently measured after grinding into powder and blended with warm water to form a consistent dough, which was then pelleted, sun-dried, packed in tight cellophane bags, and stored. The feed ingredient profile table is presented in Table 3.

Experimental diets

Based on the nutritional requirements of African catfish fingerlings [National Research Council (NRC, 2011)], five isonitrogenous and isolipidic diets were formulated at 40 % protein and 9.5 % lipids, containing 0-45% GCGM at different levels of inclusion as shown in Table 3.

Experimental conditions and fish rearing

The hybrid catfish (hetero-clarias) fingerlings, with an initial mean weight of (2.24-2.27g) were purchased from Tagwai fish hatchery of the Ministry of Livestock and Fisheries development Minna, Niger State. The fishes were transported in a well-oxygenated water plastic container from the hatchery to the old campus of the Federal University of Technology, Minna, and the feeding trial experiment was carried out at the old teaching and research farm of the Department of Water Resources, Aquaculture, and Fisheries Technology. Upon arrival, they were acclimatized in a transitional tank in the farm for four days and were fed commercial feed (Coppens feed) at 40% crude protein once a day before the experiment commenced. The fish were subsequently fed with 40% iso-nitrogenous diet and 9.5% lipid, containing different inclusion level of GCGM, designated as D1 (0% inclusion), D2 (15% inclusion), D3 (25% inclusion), D4 (35% inclusion) and D5 (45% inclusion) for 70 days. Fifteen net hapa (0.5×0.5×1m) were suspended in two outdoor concrete tanks (8mx5mx1.5m) with the aid of kuralon twine tied to plastic poles. The concrete tanks were filled to 5/6 of their volume (40m³) with filtered and dechlorinated tap water, 20 fishes were accommodated in each hapa. Each treatment was randomly allocated to three hapas, Photoperiod depends on the natural light, and the water temperature was monitored daily. The water quality parameters in the system were monitored weekly, the temperature ranged between 24-29°C while the concentration of dissolved oxygen ranged between 5.94-7.82 mg/L and the pH values of the treatments ranged from 7.18-7.90. No negative values were observed for nitrite and nitrate. Each treatment was replicated twice using 20 fish per hapa and was reared on each of the five diets. The feed was manually administered and the fish were fed to satiation three times daily at 09:00am, 12:00pm and 4:00pm. The feeding rate was subsequently adjusted according to their growth rates per hapa. The uneaten and faecal matters were siphoned out of the hapa every morning before feeding, and 45 minutes after the fish have been fed. The fish were denied feeding 24 hrs before sampling. Five fishes were randomly sampled on weekly basis, and weights were measured using a digital electronic weighing balance (CITIZEN MP-300) model.

Biochemical Analysis

About 10g initial and 15g of final samples from each hapa were pooled separately and then homogenized using

laboratory mortar and pestle. The main ingredient used for the experimental diet formulation and the body carcass of the experimental fish samples were subjected to proximate analysis. The proximate composition analysis was determined according to the Association of Official Analytical Chemists [AOAC procedures (2000)]. The moisture content analysis was carried out by oven-drying the samples at 105±3°C until a consistent weight was attained. Dried samples were used for the determination of crude fat, protein and Ash contents. Crude fat was measured by solvent extraction method in a Soxhlet system where n-hexane was used as a solvent. The crude protein value was evaluated using the Kjeldahl method. A conversion factor of 6.25 was used for the calculation of protein content according to AOAC (2000). The secondary metabolites in the raw and processed seeds; saponin, oxalate and trypsin inhibitor activity (TIA) were determined using modified AOAC (1984) procedures, while Phytic acid analysis was carried out using the modified method of Latta and Eskin (1980).

Acid insoluble ash (AIA) analysis

AIA analyses were carried out on the diets and faeces. AIA was obtained by adding 25 ml of 10% HCl to the weighed ash content of a sample. This was covered with a water glass and boiled gently over a low flame for five minutes. Ash-less filters were used to filter the solution and then rinsed with warm distilled water. The extract from the screen was taking back to the crucible and heated up until it was carbon-free and then weighed. Percentage AIA was calculated as:

$$\% \text{ AIA} = \frac{\text{Weight of AIA}}{\text{Weight of Ash}} \times 100$$

Determination of digestibility coefficient

The protein and lipid digestibility coefficient were estimated using the method according to Jimoh et al. (2010) which was evaluated based on the percentage of AIA in feed and faeces and the percentage of nutrient on diets and faeces.

$$\text{Apparent protein digestibility (\%)} = 100 - \left(\left(\frac{\text{AIA in diet (\%)}}{\text{AIA in faeces (\%)}} \right) \times \left(\frac{\text{N in faeces (\%)}}{\text{N in diet (\%)}} \right) \right) \times 100$$

Blood collection and haematological analysis

Blood samples were collected in triplicate following the procedure of Klontz and Smith (1968) and Wedemeyer and Yasutake (1977), and subsequently taken to the Laboratory of Department of Biochemistry Federal University of Technology Minna for haematological analysis. At the laboratory, the blood samples were cleaned up and prepared according to the procedure described by Ogbu and Okechukwu (2001). The Packed cell volume (PCV), Haemoglobin (Hb), and Red blood cell (RBC) were measured directly and total erythrocyte indices (MCH, MCV

Table 2. Effect of treatment on anti-nutritional factors of *Canavalia gladiata* seed meal (GCGM).

Anti-nutritive factors	RCGM	GCGM	(%) decrease of anti-nutritive factors after germination
Phytate (mg/g)	21.55	5.34	75.22
Oxalate(mg/g)	2.85	0.18	93.68
Tannin (mg/g)	0.05	0.01	80.00
Saponin (g/100g)	5.50	0.88	84.00

^aMeans of two replicate analyses.

Key: **RCGM** = Roasted *Canavalia gladiata* Seed Meal; **GCGM** = Germinated *Canavalia gladiata* Seed Meal.

and MCHC) were calculated. The white blood cell was analyzed as described by Dacie and Lewis (2001).

$$MCV = \frac{PCV}{\text{Erythrocytes count}} \times 10$$

$$MCH = \frac{\text{Haemoglobin}}{\text{Erythrocytes count}} \times 10$$

$$MCHC = \frac{\text{Haemoglobin}}{PCV} \times 100$$

Evaluation of Nutrient Utilization Parameters

Nutrient Utilization was analyzed in terms of Feed Efficiency (FE), Specific Growth Rate (SGR), Feed Intake (FI), Protein Efficiency Ratio (PER) and Protein Retention (PR).

The following formulas were used:

$$\text{Weight gain (\%)} = \frac{(\text{Final weight (g)} - \text{initial weight (g)})}{\text{initial weight (g)}} \times 100$$

$$\text{Feed efficiency (\%)} = \left(\frac{\text{weight gained (g)}}{\text{feed fed (g)}} \right) \times 100$$

$$\text{Specific growth rate (\%)} = \left(\frac{\text{In final weight (g)} - \text{In initial weight (g)}}{\text{feeding period (day)}} \right) \times 100$$

$$\text{Feed intake (mg/fish/day)} = \frac{\text{dry feed (mg) given / number of fish}}{\text{feeding period (day)}}$$

$$\text{Protein efficiency ratio} = \frac{\text{weight gain}}{\text{protein intake (g)}}$$

$$\text{Protein retention (\%)} = \frac{\text{protein gain}}{\text{protein fed}} \times 100$$

Statistical analyses

Data were analyzed using one-way analysis of variance (ANOVA) using Minitab 17 (Stat-Soft, Inc., Oklahoma, USA). Differences between treatments were compared by Tukey's test. The significance level was tested at $P < 0.05$.

RESULTS AND DISCUSSION

The proximate compositions of the experimental ingredients (Table 1) revealed that crude protein is one of the major constituents of the *Canavalia gladiata* seed. The crude protein content value of RCGM was 28.61%, this value is

within range and compares well with earlier reports on sword bean (Bressani et al., 1987; Revilleza et al., 1990; Vadivel and Janardhanan, 2004). However, the crude protein value of germinated *Canavalia gladiata* was higher than crude protein (35.82 %) (Table 1). The higher crude protein obtained in germinated *Canavalia gladiata* meal (GCGM) could be attributed to the processing method to which the seeds were subjected to (germination). This increase in proteins may be due to loss of dry weight as some fats and carbohydrates are utilized during respiration whereas some amino acids are synthesized during germination (Onyango et al., 2013; Jan et al., 2017). Germination is used in processing many plant ingredients to improve and boost their nutritional quality as it results in the reduction of anti-nutritional factors furthermore germination increased the amount of riboflavin, thiamin, ascorbic acid and niacin and also boost the protein content of leguminous plant seeds (Abdullah and Baldwin, 1984; Kylene and McCready, 1975; Fordham et al., 1975; Onyango et al., 2013; Oghbaei and Prakash, 2016). However, Laxmi et al. (2015) reported that an increase in protein content upon germination depends on the type of the plant.

Table 2 showed that there was a significant reduction in the value of the anti-nutritional components (phytate, oxalate, tannin, saponin) present in the RCGM after subjecting *Canavalia gladiata* to germination. In raw *Canavalia gladiata*, the phytate had higher (21.55) and tannin had the lowest value (0.05). Phytate has been considered an anti-nutritional factor because it interacts with food constituents such as minerals and makes them unavailable (Idris et al., 2006). Similarly, Abdel-Rahaman et al. (2007) reported that germination of various pearl millet cultivars significantly reduced the phytic acid and polyphenol contents of the cultivars. The decrease in the anti-nutritional factor (phytate) could be attributed to phytate activity in the germinating grains (Larsson and Sandberg 1992; Sokrab et al., 2012). The effect of germination on tannin shows a significant reduction. A similar trend was obtained by Singh et al. (2014) for pulses; Vijayakumar et al. (1998) for *Vigna aconitifolia* and *Vigna sinensis* and Jood et al. (1987) for genus *Canavalia*. Nkhata et al. (2018) reported that the germination

Table 3. Formulation profile and proximate composition of experimental diets (g/kg).

Ingredients	D1	D2	D3	D4	D5
Fish meal	612.70	524.20	465.90	407.40	349.00
Soybean meal	50.00	50.00	50.00	50.00	50.00
GCGM	0.00	150.00	250.00	350.00	450.00
Yellow maize meal	25.00	25.00	25.00	25.00	25.00
Millet	25.00	25.00	25.00	25.00	25.00
Starch	50.00	50.00	50.00	50.00	50.00
Vitamin premix	15.00	15.00	15.00	15.00	15.00
SBO	19.30	15.60	12.70	10.10	7.60
Mineral	15.00	15.00	15.00	15.00	15.00
Cellulose	188.00	130.20	91.40	52.50	13.40
Total	1000.00	1000.00	1000.00	1000.00	1000.00
Moisture (%)	4.83	4.65	4.71	4.81	4.48
Crude protein (% d.b.*1)	37.54	37.42	37.35	37.43	37.48
Crude lipid (% d.b.*1)	8.58	8.76	8.62	8.84	8.87
Ash (% d.b.*1)	9.28	9.36	9.58	9.62	9.57
Crude fibre (% d.b.*1)	6.64	6.54	6.44	6.38	6.68
AIA (% d.b.*1)	4.28	4.30	4.26	4.42	4.58

FM = Fish meal; **SBM** = Soybean meal; **GCGM** = Germinated *Canavalia gladiata* Seed Meal; **MM** = Yellow maize meal; **SBO** = Sheabutter oil; **d.b.*1** = dry basis; **AIA** = Ash insoluble Ash.

****:** Premix composition: vitamin and mineral premix (IU or mg / kg of premix). Vitamin A: 4800 IU; Cholecalciferol (vitamin D): 2400 IU; Vitamin E: 4000 mg; Vitamin K: 800 mg; Vitamin B1: 400mg; Riboflavin: 1600 mg; Vitamin B6: 600 mg, Vitamin B12: 4 mg; Pantothenic acid: 4000 mg; Nicotinic acid: 8000mg; Folic acid: 400 mg; Biotin: 20 mg, Manganese: 22000 mg; Zinc: 22000 mg; Iron: 12000 mg; Copper: 4000 mg; Iodine: 400 mg; Selenium: 400mg; cobalt: 4.8 mg.

processing technique is commonly used to disrupt the interaction of anti-nutritional factors and make nutrients and phytochemicals free and accessible for digestive enzymes. This is associated with the activation of some endogenous enzymes making germinated *Canavalia gladiata* higher in nutritional quality compared to raw *Canavalia gladiata* meal (Sridhar and Seena, 2006; Zhang et al., 2015). GSBM had a 75.22% reduction in Phytate, 93.68% Oxalate, 80.00% Tannins and 84.00% Saponin. Germination has proven to be useful in removing/reducing certain unwanted heat-stable components like phytates and tannins. The hydrolyzation of phytic acid in beans during germination was reported by (Reddy et al., 1982). The breakdown of phytate during germination can be attributed to the increased activity of the endogenous phytase (enzyme activity) (Shimelis and Rakshit, 2007). Furthermore, it has been observed that reduction in tannin content may be attributed to the formation of hydrophobic association of tannins with seed proteins and enzymes as reported by (Sharma and Sehgal, 1992) in sprouted bean seeds.

Fish mortality was low and relatively uniform in all the treatments. Furthermore, all the experimental fish remaining

in the tanks were morphologically normal at the end of the feed trial. There were no significant differences in the survival rate amongst the treatment means. The survival was splendid and this could be attributed to proper handling and good water quality parameters maintained during the experimental period.

Sogbesan and Ugwumba (2008) reported that adequate and right processing of ingredients has an effect on the texture and palatability of the experimental diets. The values for growth indices performance WG, FWG and FI were significantly high in fish fed diet D4 which suggest the breakdown of amino acids and improve the digestibility of feed protein as reported by (Shimelis and Rakshit, 2007). The result obtained in growth parameters showed significant differences amongst the treatment means. The D4 had the highest value (977.1±16.2) while D5 had the lowest (841.7±22.2) value in terms of weight gain (Table 4). However, there was no significant difference between D1, D2, D3 and D5 respectively. The weight gain increased as the inclusion levels increases from 0 – 35 %, though no improvement obtained in weight gain at 45% level as compared to the control diet D1. Adewole and Olaley (2014) reported

Table 4. Growth performances and nutrient utilization of hybrid catfish fingerling fed experimental diets for 70 days.

Diet code	Body weight (g)		Weight gain (%)	Survival rate (%)	Total feed intake (g)	Feed efficiency	Protein efficiency ratio	Protein retention (%)
	Initial	Final						
D1	2.27±0.44	21.68±0.61 ^b	854.92±12.36 ^b	99.60±2.34 ^a	24.62±0.48 ^{ab}	0.79±0.57 ^b	2.10±0.37 ^b	40.46±0.42 ^b
D2	2.24±0.34	21.65±0.73 ^b	866.35±15.23 ^b	99.72±2.48 ^a	24.69±0.26 ^{ab}	0.79±0.71 ^b	2.10±0.15 ^b	40.44±0.38 ^b
D3	2.24±0.52	22.04±0.66 ^b	884.14±18.42 ^b	99.76±2.39 ^a	25.96±0.57 ^{ab}	0.79±0.35 ^b	2.10±0.33 ^b	40.48±0.51 ^b
D4	2.26±0.28	24.38±0.34 ^a	977.07±16.24 ^a	99.80±2.55 ^a	26.96±0.44 ^a	0.82±0.42 ^a	2.19±0.16 ^a	42.05±0.14 ^a
D5	2.55±0.36	21.39±0.72 ^b	841.65±22.16 ^b	99.68±2.34 ^a	24.46±0.76 ^b	0.78±0.53 ^c	2.09±0.12 ^c	40.25±0.24 ^c

Values in the same column with different superscript letters are significantly different ($p < 0.05$) from each other.

Table 5. Apparent digestibility coefficient of *Hybrid catfish* fingerlings fed experimental diets for 70 days.

Diet code	ADC of crude protein (%)	ADC of crude lipid (%)	ADC of crude fibre (%)
D1	87.3±1.4 ^a	82.0±1.2 ^a	52.1±1.6 ^b
D2	87.3±1.3 ^a	82.3±1.1 ^a	53.4±1.3 ^b
D3	87.6±1.5 ^a	82.3±2.4 ^a	52.3±1.5 ^b
D4	87.2±1.4 ^a	82.1±1.5 ^a	53.2±1.3 ^b
D5	86.8±2.5 ^b	81.9±1.2 ^a	54.4±1.2 ^a

^a Means of two replicate analyses

that the weight gain of fingerlings is usually a reliable indicator of the nutritional adequacy of the diet. There were no significant differences between D1, D2, and D3; however, D4 was significantly higher and different in total feed intake than D5 (Table 4).

Factors such as water, temperature, fish size, feeding frequency, photoperiod, stocking density and feed quality are known to modify the feed intake in fish (Eriegha and Ekokotu, 2017). However, Singh et al. (2003) reported that the response of fish for feed varies among the species. The protein efficiency ratio and protein retention followed the same pattern and varies significantly amongst treatment means, Fish fed D4 had the highest significant PER value while fish fed D5 diet had the lowest value and was significantly lower than other fish fed experimental diets, though there were no significant differences between D1, D2, and D3. This result is in accordance with the findings of Ndako et al. (2019). Kiriratnikom and Kiriratnikom (2012) also reported that at higher temperature, the catfish required higher dietary protein level to maximize protein retention.

The apparent digestibility coefficient of the experimental diets is presented in Table 5. The crude fibre value of D5 was exceptionally and significantly ($P < 0.05$), higher than other formulated experimental diets (D1, D2, D3 and D4). However, there was no significant difference ($P > 0.05$) between D1, D2, D3 and D4. Furthermore, there was no significant difference ($P > 0.05$) in the apparent digestibility coefficient

crude lipid of all the formulated experimental diets fed D1, D2, D3, D4 and D5, while the crude protein digestibility value of D5 was significantly lower than the other formulated experimental diets.

The carcass compositions of the experimental fish revealed that the carcass protein content was not significantly different (Table 6). However, they were significantly higher compared to the initial (before the experiment). This indicates that the fish were able to retain nutrient in their body as they feed on experimental diets. Wu et al. (2014) posited that protein contributes to a wide range of functions in muscular tissues. The diet protein that could not be retained by cultured fish causes various environmental problems including eutrophication and disease outbreak (Rahman, 2015). The moisture content of the experimental fish was significantly lower compared with the initial (Table 6). The qualitative determination of fish moisture content is paramount in any quality control programme (Nurullah et al., 2007). Olagunju et al. (2012) reported that the moisture content of the fish sample is an indication of the wetness caused by water and could also be due to stable water levels in the environmental location. The lipid contents were significantly different, though D4 and D5 were not different significantly (Table 6). However, they were higher compared to the initial value. Alasalvar et al. (2002) reported that lipid from fish is well known as a rich source of some long-chain n-3 polyunsaturated fatty acids which cannot be synthesized

Table 6. Proximate composition analyses of whole-body hybrid catfish (wet basis) fed experimental diets for 70 days.

Component (%)	Initial	Final*1				
		D1	D2	D3	D4	D5
Moisture	77.6	75.2±0.3 ^a	74.1±0.6 ^b	73.22±0.8 ^c	72.67±0.5 ^d	72.15±0.7 ^d
Protein	13.5	18.7±1.3	18.7±1.5	18.7±1.7	18.6±1.4	18.6±1.6
Lipid	4.1	4.2±0.4 ^c	4.5±0.2 ^c	5.2±0.6 ^b	6.2±0.4 ^a	6.8±0.5 ^a
Ash	2.0	2.2±0.1	2.3±0.3	2.4±0.1	2.4±0.3	2.3±0.2

*1 Values in the same row with different superscript letters are significantly different ($p < 0.05$) from each other ($n = 3$).

Table 7. Haematological parameters of Hybrid catfish fingerling fed experimental diets for 70 days.

Blood Parameter	Initial	Final*1				
		D1	D2	D3	D4	D5
PCV (%)	20.24	32.56±1.52 ^a	32.54±0.45 ^a	32.57±0.68 ^a	32.58±0.74 ^a	32.36±0.33 ^a
WBC (10^3 mm^{-3})	5.46	6.14±0.61 ^a	6.20±0.18 ^a	6.30±0.23 ^a	6.43±0.55 ^a	6.86±0.79 ^a
RBC (10^3 mm^{-3})	1.98	3.17±0.05 ^a	3.17±0.82 ^a	3.15±0.26 ^a	3.14±0.61 ^a	3.13±0.28 ^a
Hb (g/100 ml)	6.12	9.94±0.57 ^a	9.88±0.46 ^a	9.80±0.73 ^a	9.82±0.65 ^a	9.72±0.42 ^a
LYMPH (100)	60.38	61.40±0.72 ^a	61.59±0.58 ^a	61.78±0.62 ^a	61.85±0.77 ^a	61.66±0.35 ^a
MCHC (%)	30.24	30.53±0.37 ^a	30.36±0.28 ^a	30.09±0.42 ^a	30.14±0.56 ^a	30.04±0.74 ^a
MCH (pg)	30.91	31.36±0.24 ^a	31.17±0.46 ^a	31.11±0.62 ^a	31.27±0.31 ^a	31.05±0.52 ^a
MCV (fl)	102.22	102.70±0.64 ^a	102.65±0.48 ^a	103.40±0.24 ^a	103.76±0.75 ^a	103.39±0.18 ^a

PCV, packed cell volume; WBC, white blood cell; RBC, red blood cell; Hb, haemoglobin; LYMPH, lymphocyte; MCHC, mean corpuscular haemoglobin concentration; MCH, mean corpuscular haemoglobin concentration; MCH, mean corpuscular haemoglobin; MCV, mean corpuscular volume.

*1 Values in the same row with different superscript letters are significantly different ($p < 0.05$) from each other ($n = 3$).

by humans and commonly from the diet. The lipid content is directly related to the nutrition of the fish (Begum et al., 2012).

The haematological study of cultured species is a significant instrument in the development of the aquaculture system, particularly in the detection of healthy from diseased/stressed animal (Rainza-Paiva, et al., 2000). There were no significant differences amongst the treatment means in white blood cell (WBC) between D1, D2, D3, and D4 while D5 was significantly different from others (Table 7). However, they were higher when compared with the initial value. This could be attributed to the activation of the defence mechanism. Akinwande et al. (2004) reported that a measurable increase in the WBC count of fish is a function of immunity and animal resistance to some vulnerable diseases. The packed cell volume (PCV) values were not significantly different, though D5 differed significantly from others (Table 7). However, the values obtained were significantly higher, when compared with the initial. PCV is a

reliable indicator of various sources of stress leading to decreased fish activeness (Satheesh et al., 2011).

CONCLUSION

From the result obtained in this study, it is concluded that fingerlings of *Clarias gariepinus* can make use of germinated sword bean seed meal at 35% in their diets to give excellent performance in growth, nutrient utilization and body composition without any adverse effect on their health. However, an increase in the inclusion level above 35 % would lead to depression in their growth response. Furthermore, for effective utilization of germinated sword bean seed meal, there is a need to evaluate the environmental impact of this sword bean seed meal-based diet in a long-term pond culture system. Therefore, further research on this aspect of digestion and mechanism of amino acid and fatty acid metabolism of the germinated sword bean meal in fish feed diet needs to be evaluated.

Conflict of Interests

The author(s) declare no conflict of interest.

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