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Research Article

Growth Performance and Economic Evaluation of *Clarias gariepinus* Fingerlings Fed Fermented *Canavalia ensiformis* Seed Meals

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ABSTRACT

Fish farms' output and profits have been facing a greater challenge with a hike in the price of feed ingredients. This study was designed to evaluate the growth and economic performance of *clarias gariepinus* fingerlings fed on fermented *Canavalia ensiformis* seed meals. The proximate analysis of the diets was done according to Jiyana, *et al*. (2022) method. Fermented *Canavalia ensiformis* meals were used to formulate five different 40.0% isoproteic experimental diets each at 0%, 25%, 50%, 75% and 100% and coded FCe, which were fed at 5% body weight to fingerlings of *Clarias gariepinus* fingerlings. One hundred and fifty *Clarias gariepinus* (0.96g±0.03 and 3.93cm±0.28) were randomly assigned to five treatments of 10 fingerlings, replicated three times in a completely randomised design (CRD) in 35L plastic circular tanks through a semi-flow-through system for 84days. Weights and lengths were measured biweekly and used to calculate the growth and nutrient utilization parameters. Nutrients, digestibility, haematology, and economic evaluation of the fish fed the diets were also determined. Data collected were analysed using a one-way analysis of variance. The results of the experiments showed that there were significant differences (p<0.001) in the specific growth rate as the inclusion levels of the tested meals increased. The mean water quality parameters were within the tolerable limit for aquaculture. Nutrient digestibility and Haematology were significantly (p<0.001) reduced in the treatment diets. Inclusion of the tested meals in the diets resulted in the reduction of feed cost/Kg, cost of feed intake/fish and feed cost/weight gain with increased inclusion levels. *Clarias gariepinus* fingerlings can feed on up to 25% inclusion level fermented *Canavalia ensiformis* diet without adverse effects on the growth performance.

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INTRODUCTION

The recent increase in aquaculture development in Nigeria and the world at large is accompanied by the need to be incorporating plant materials in fish feed (Hardy, 2010), to dispense total reliance on conventional legumes that have successfully replaced fishmeal. Soybean (Glycine max) which could replace not less than 50% of the fishmeal in the diet of fish species is not much available in the market and commands a high price because of its competitive use as food by man and feed ingredients for livestock feed producers (Siddhuraju and Becker, 2001), and hence the need to shift priority to searching for alternative plant protein feed ingredients of little or no use and cheaper in terms of cost to replace these conventional legumes. The search for alternative feedstuff to the current conventional feed ingredients requires that the digestibility of the alternative feed ingredients be favourably compared to the conventional feed ingredients (Sarker *et al*., 2016).

Legumes have been recognized to be the second most valuable plant source for human and animal nutrition and the third largest family among flowering plants, consisting of approximately 650 genera and 20000 species (Sridhar *et al*., 2007; Wojciechowksi *et al*., 2004). Kalidass *et al*. 2012 reported that legume seeds are important sources of nutrients and can serve as high-quality dietary protein to meet the nutrient requirements of fish (Perumal *et al*., 2001; Escudero *et al*., 2006).

The wild legumes, which have tremendous potential for commercial exploitation but remain ignored, form a good scope in this context (Arivalagan *et al*., 2014). Inbasekar, (2014) reported that common proteinaceous edible legumes (soybean (Glycine max) and cowpea (*Vigna unguiculata*) are available in the market, and in most cases, production rates are compared with consumption (as food and feed) have remained un-met, and an ever-increasing demand has been witnessed by food and feed industries. Additionally, an unjustified shortage of plant protein resources has resulted in the majority of the world's population transitioning from an animal-based diet to one based on a protein-rich vegetarian diet. In this regard, the use of legumes as a successful alternative to animal products has been highlighted (Famurewa, 2005).

Canavalia ensiformis; belongs to the Kingdom- Plantae, Subkingdom- Viridiplantae, Division- Tracheophyta, Subdivision- Spermatophytina, Class- Magnoliopsida, Order - Fabales, Family – Fabaceae, Genus – Canavalia DC. Jackbean and Species – Canavalia ensiformis (L) DC. – wonderbean. The genus *Canavalia* comprising of 48 species of these underutilized legumes (Fagbenro *et al*., 2007). *Canavalia ensiformis* is an annual or weak perennial legume with climbing or bushy growth forms. It is woody with a long taproot. The 8 in (20 cm) long and 4 in (10 cm) wide leaves have three egg-shaped leaflets that are wedge-shaped at the base, and taper towards the tip. The 1 in (2.5 cm) long flowers are rose-coloured, purplish, or white with a red base. It has a 12 in (30 cm) long, 1.5 in (3.8 cm) wide, sword-shaped seed pod. Seeds are white, red, brown and smooth with a brown seed scar that is about one-third the length of the seed. Its roots have nodules which fix Nitrogen (Fagbenro, *et al*., 2007; Tiamiyu *et al*., 2016). They are widely distributed and indigenous to the tropics (Fagbenro, *et al*., 2007), rarely eaten by man (Solomon, *et al*. 2017) and their nutritional potential has been well studied in the Monogastric (Fagbenro, *et al*., 2013 and Doss, *et al*., 2011). Nutritional trial in fish includes the works of the previous authors (Akande, *et al*., 2010; Audu, *et al*., 2011;

Cheng, *et al*., 2003; Okomoda, *et al*., 2006; Fagbenro *et al*., 2007).

Alonso *et al*. (2000) define anti-nutritional factors (ANFs) as innate components of a food/feed ingredient that have a limiting effect on food/feed intake, digestion, and nutrient absorption. Possibly the most limiting factor for the use of plant feed ingredients as nutrient sources for fish are ANFs inherent to them. Francis *et al.* (2001) reported that ANFs in legumes can be divided into several groups based on their chemical and physical properties such as non-protein amino acids, quinolizidine alkaloids, cyanogenic glycosides, pyrimidine glycosides, isoflavones, tannins, oligosaccharides, saponins, phytates, and lectins or protease inhibition. Their elimination can be achieved either by a selection of plant genotypes with low levels of such factors or through post-harvest processing (germination, cooking, boiling, leaching/soaking, soaking and toasting, fermentation). This study intends to use fermentation methods to eliminate the anti-nutrients in *Canavalia ensiformis* seed meals and its inclusion level effects on the growth of *Clarias gariepinus*.

MATERIALS AND METHODS

Study area

The feeding trial was conducted at the Fisheries Research Farm of the Department of Fisheries, Moddibo Adama University of Technology (MAUTECH), Yola. Adamawa State is located at latitude 9.140N, longitude 12.380E and an altitude of 185.9m. Girei is located on latitude 9.220N, longitude 12.330E an altitude of 245m. It has an average annual rainfall of about 759mm with a maximum temperature of 39.70C. The rainy season run from May through October, while the dry season commences in November and ends in April. The driest months of the year are January and February when the relative humidity drops to 13% (Pandey, 2003; Louis *et al*., 2018).

Canavalia ensiformis fruits were collected from Girei and their surroundings in Adamawa State. They were identified using a field handbook by Roque *et al.* (2012), a plant Taxonomist at the Forestry and Wildlife Department of Modibbo Adama University of Technology, Yola (MAUTECH). Raw seeds were moistened with water, kept in a container with a cover to ferment for 72 hours under laboratory conditions, oven-dried at $50^{o}C$ then milled and tagged fermented seed meal (FSM) (Doss *et al*., 2011).

Determination of Nutrient Compositions

The seed meals were analyzed for nutrient compositions (Jiyana *et al*., 2022).

Feed Formulation

A basal diet of 40% crude protein was formulated from the commercial ingredients (fish meal, *Glycine max* meal, yellow maize, Vitamin, Minerals, Palm oil, dicalcium phosphate and starch), and *Glycine max* (3kg) was toasted. The dry ingredients were milled with a grinding machine (Aarti M-0. Working Surface of the table (225 x 450 mm) to very fine particle size and sieved with 0.2mm sieve. The ingredients were weighed and mixed to homogeneity and pelleted with a pelleting machine with a 0.2 mm diameter size. The pellets were air-dried at room temperature and stored in a refrigerator (with a shelf life of 2 months at temp. 25-30^{o}C) until the commencement of the feeding. Fermented *Canavalia ensiformis* were included in the experimental diets to replace *Glycine max* meal as a plant protein source at inclusion levels of 0% (control), 25%, 50%, 75% and 100% as shown in Table 1.

Table 1: Percentage Compositions of Ingredients with Fermented *Canavalia ensiformis* Meal Inclusion levels

Ingredients (g/100g)	Control (0%)	FCe (25%)	FCe (50%)	FCe (75%)	FCe (100%)
Fishmeal (68%)	32.00	32.00	32.00	32.00	32.00
Glycine max meal (46%)	33.00	24.75	16.50	8.26	0.00
Fermented *Canavalia ensiformis* (34.7%)	0.0	10.93	21.87	32.81	43.75
Yellow maize (10)	30.00	27.32	24.63	19.29	19.25
*Vitamin/mineral premix	1.0	1.0	1.0	1.0	1.0
Lysine	0.5	0.5	0.5	0.5	0.5
Methionine	0.5	0.5	0.5	0.5	0.5
Palm oil	1.0	1.0	1.0	1.0	1.0
Cassava starch	1.0	1.0	1.0	1.0	1.0
Dicalcium phosphate	0.5	0.5	0.5	0.5	0.5
Common salt	0.5	0.5	0.5	0.5	0.5
Total	100.0	100.0	100.0	100.0	100.0
Calculated Crude protein	39.94	39.67	39.40	38.87	38.87
**Gross Energy (KJ/100g)	1868.60	1818.80	1769.70	1766.40	1732.90
GE:CP	46.78	45.84	44.91	45.13	44.58

FCeM – Fermented *Canavalia ensiformis* meal

GE: CP= gross energy: crude protein

**Calculated Gross Energy (KJ/100g) = Protein x 23.6KJ/100g + Lipid x 39.5KJ/100g + NFE x 17.2KJ/100g (Blaxter, 1989)

*Vitamin-Mineral premix provides per kg the following: 12.000.000 IU Vitamin A; 2.000.000 IU Vitamin D3; 10g Vitamin E; 2g Vitamin K3; 1g Vitamin B1; 5g Vitamin B2; 1.5 g Vitamin B6; 10g Vitamin B12; 30g Nicotinic acid; 10g Pantothenic acid; 1g Folic acid; 50g Biotin; 250g Choline chloride 50%; 30g Iron; 10g copper; 50g Zinc; 60g Manganese; 1g Iodine; 0.1g Selenium and Cobalt 0.1g.

Collection and Acclimatization of Experimental Fish

One hundred and fifty (150) fingerlings of *Clarias gariepinus* were obtained from Aqua Guide Farm in Federal Housing Estate, Girei LGA, Adamawa State, Nigeria and were used for the feeding trials. The fish were kept to acclimatize to the farm conditions (24 hours) and fed with the experimental diets for twelve weeks (Tiamiyu *et al*., 2016)

Experimental Design

A completely randomized design was used, where one hundred and fifty fingerlings were randomly allocated to 5 experimental groups (10 fingerlings of *Clarias gariepinus* per bowl and replicated).

Data Collections

Growth Performance and Nutrient Utilization

The initial weight and length of fish in each treatment were taken at two weeks' intervals. The feed rations fed were adjusted based on the new weight. The weight and length recorded were used to determine the growth performance of the fish and the feed supplied was also used to determine the nutrient utilization parameters following the methods of Udo and Umoren (2011).

Growth Performance (GP)

At the end of 12 weeks, the growth rates (GR), condition factor (K), survival rate

(SR) and nutrient utilization (NU) were computed and analyzed.

Weight gain (g)

The total and mean weight gains were calculated for each replicate and treatment as follows

$$\text{Weight gain / fish (g/fish)} = (W_f - W_I)$$

$$\text{Mean weekly weight gain (g/week)} = \frac{W_f - W_I}{n}$$

Where:

W_f =final weight of fish at the end of the experiment

W_I=Initial weight of fish at the beginning of the experiment

n=Number of weeks.

Relative growth rate (RGR)

This is the percentage ratio of the weight gain to the initial body weight and was determined as follows:

$$RGR\ (\%) = \frac{W_f - W_I}{W_I} \times 100$$

Specific growth rate (SGR %/day)

This is the percentage of daily weight gain and was computed according to the formula below:

$$SGR\ (\%/day) = \frac{\text{Log } W_f - \text{Log } W_I}{t} \times 100$$

Where:

Log W_f = Logarithm of the fish final weight

Log W_I = Logarithm of the fish's initial weight

t = Experimental period in days

Condition factor (K)

This expresses the health status of fish as a result of the experimental treatment and was computed at the beginning and end of the experiment (K1 and K2) using Fulton's condition factor formula as expressed by Udo and Umoren, 2011 as:

$$K = \frac{100W}{L^3}$$

Where:

W = Weight of fish

L = Length of fish

Survival Rate (%)

$$SR\ (\%) = \frac{N_f}{N_i} \times 100$$

Where:

N_i = Number of cultured fish stocked at the beginning of the experiment

N_f = Number of fish alive at the end of the experiment

Feed Utilization

Feed conversion ratio (FCR)

This is a numerical value used to measure the utilization of feed for growth. The feed conversion ratio was calculated according to Udo, I.U. and Umoren, U.E. (2011) method.

$$FCR = \frac{\text{Feed Intake}}{\text{Weight gain (g)}}$$

Feed intake (FI)

This will be taken as the addition of the amount of feed supplied during the experimental period.

Protein intake (PI)

This is the numerical value of the quantity of protein present in the feed that was fed to the fishes during the experimental period and was determined as follows, Getso, *et al*., 2017:

Protein Intake = Feed Intake x Crude protein

Protein efficiency ratio (PER)

This index uses growth as a measure of the nutritive value of dietary protein as:

$$PER = \frac{\text{Mean weight gain (g)}}{\text{Mean Protein Intake (g of protein in 100g of diet/fish)}}$$

Water Quality Parameters

Water quality parameters such as Temperature, pH, Dissolved Oxygen and Ammonia concentration were taken biweekly before feeding using the NIFFR (National institute for freshwater fisheries research, new bussa, Niger state, Nigeria) innovative multitec kit (2010) and Ammonia by titration with sulfuric acid.

Determination of Nutrient Digestibility Coefficient

Indirect methods of digestibility were applied by using Chromium III Oxide incorporated into experimental diets as an inert indicator of digestion (Fagbenro, *et al*., 2007). Faecal samples were collected twice a week at regular intervals by slightly pressing the anal region of the fish after seven hours of feeding. The samples that were collected are weighed, ground in a mortar, sealed in polythene bags, labelled (according to each dietary treatment code) and kept in a refrigerator before proximate analysis. The indices were calculated as:

Apparent digestibility of dry matter (ADdm)

$$ADdm\ (\%) = \frac{(1 - \text{dietary Chromic oxide})}{\text{Feacal chromic oxide}} \times \frac{\text{Feacal dry matter}}{\text{dietary dry matter}} \times 100$$

Apparent digestibility of protein (ADp)

$$ADp\ (\%) = \frac{(1 - \text{dietary chromic oxide})}{\text{Feacal chromic oxide}} \times \frac{\text{feacal protein}}{\text{dietary protein}} \times 100$$

Apparent digestibility of energy (ADe)

$$ADe\ (\%) = \frac{(1 - \text{dietary chromic oxide})}{\text{Feacal chromic oxide}} \times \frac{\text{feacal protein}}{\text{dietary energy}} \times 100$$

Haematological Examination

Collection of blood sample

At the end of the feeding trials, three samples of live fish from each treatment were removed according to Dacie and Lewis (1977) method. 10 ml of blood was collected from the caudal peduncle of each fish using a separate heparinized 10ml disposable syringe, kept into properly labeled sterilized bottles (EDTA-01A-500) containing EDTA (Ethylene diamine tetra-acetic acid) as anticoagulant and transported to the laboratory for analysis according to Sveier *et al*. (2000) methods.

Haematocrit (PCV)

Heparinized capillary tubes were 75% filled with blood samples by suction pressure and one end sealed with plasticine. The tubes were centrifuged for 5 minutes in a haematocrit centrifuge (CFG-12D) at 3000 r.p.m. The packed cell volumes (PCV) were read by the use of a haematocrit reader. The results were expressed in percentages.

Haemoglobin concentration (Hb)

The cyanmethemoglobin method by Sveier *et al*. (2000) was used. 0.02ml of well-mixed blood was added to 4ml of modified Drabkins solution (a mixture of 250mg potassium ferricyanide, 200mg potassium cyanide and 50mg of potassium dihydrogen phosphate) and the volume was diluted to 1 litre with distilled water. The mixture was allowed to stand for 35 minutes and the Haemoglobin concentration (g/dl^{-1}) was read photometrically by comparing it with the cyanmethemoglobin standard with the yellow-green filler at 625nm.

Leucocyte count (WBC)

Haemocytometer was used for LC determination with a 0.8cm objective of the microscope and large squares (area = $1mm^2$, depth = 0.1mm) the volume was $0.1mm^3$ and the dilution factor was 20. Four squares were used and the total counts per mm3 were:

20 x 1 x L cells ÷ 0.4 = 50 x L cells

Where: L = number of leucocytes counted

Erythrocytes: Red Blood Cells (RBC)

These were determined in heparinized blood diluted by Hayman solution at a ratio of 1:200. Neubauer improved haemocytometer placed on a compound Olympus Microscope (BX51) stage was used to count/estimate the erythrocyte population. The number of cells counted, R (average of two fields) was multiplied by the dilution factor and the volume of $1/4000mm^3$ (area = $1/4000mm^3$, depth = $1/10mm^3$) and counting were done in 80squares with the total volume of $1/50mm^3$ the dilution factors is 200.

200 x 500 x R cells = 10,000 x R

Determination of Mean Corpuscular Haemoglobin (MCH)

The mean corpuscular haemoglobin (MCH) was calculated using the formula

$$\text{MCH (pg)} = \frac{\text{Hb x 10}}{\text{RBC}}$$

Determination of Mean Corpuscular Volume (MCV)

The mean corpuscular volume (MCV) was determined as

$$\text{MCV (fL)} = \frac{\text{Packed cell volume x 10}}{\text{RBC}}$$

Determination of Mean Corpuscular Haemoglobin Concentration (MCHC)

The mean corpuscular haemoglobin concentration (MCHC) was calculated using the formula

$$\text{MCHC (g/dL)} = \frac{\text{Hb x 100}}{\text{PCV}}$$

ECONOMIC ANALYSIS

The economic analyses were computed to estimate the cost of feed required to raise a kilogram of fish using the various experimental diets. The major assumption is that all other operating costs for commercial fish production will remain the same for all diets. Thus, the cost of feed was the only economic criterion in this case. The cost was based on the current prices of the feed ingredients at the time of purchase. The economic evaluations of preparing the diets were calculated from the method of Witeska , *et al*. (2022) and Agbo *et al*. (2015).

Investment Cost Analysis (ICA) = Cost of Feeding (₦) + Cost of Fingerlings stocked (₦)

Profit Index (PI) =Net profit value (₦) / Cost of feeding (₦)

Incidence of Cost (r) = Cost of feeding (₦) / Weight of fish produced (g).

Benefit- Cost Ratio (BCR) =Net profit value (₦) / Investment Cost Analysis (₦)

Statistical Analysis

Data collected were subjected to Descriptive statistics, graphical representations (Bar charts) and one-way analysis of variance ANOVA. Significant differences between treatment means were compared using the least significant difference (LSD) at a 5% probability level using IBM SPSS statistics 20.

Proximate Compositions of Fermented *Canavalia ensiformis* (FCe) Diets

The highest crude protein value was from the control diet (40.36%) and the lowest from the 100% FCe diet (39.40%). The highest lipid content was from the control diet (7.01%) and the lowest from the 25% FCe diet (6.75%). The highest fibre value was from the 100% FCe diet (5.76%) and the lowest from the control diet (5.43%). The Highest Ash content was from the 100% FCe diet (6.60%) and the lowest was from the control diet (6.52%). The highest nitrogen-free extract was from the 100% FCe diet (32.77%) and the lowest was from the control diet (32.18%). The highest dry matter content was from the 75% FCe diet (91.51%) and the lowest was from the 50% FCe diet (91.40%). The highest gross energy value was from the control diet (417.71Kcal/g) and the lowest was from the 100% FCe diet (414.48Kcal/g) in Table 2.

RESULTS

Table 2: Proximate Compositions of Fermented *Canavalia ensiformis* Diets on Dry Matter Basis

Nutrients	**FCe** 0%	**FCe** 25%	**FCe** 50%	**FCe** 75%	**FCe** 100%
Protein %	40.36	40.25	40.15	39.45	39.40
Fat %	7.01	6.75	6.84	6.95	6.97
Fibre %	5.43	5.66	5.72	5.75	5.76
Ash %	6.52	6.55	6.55	6.57	6.60
NFE %	32.18	32.24	32.14	32.79	32.77
Dry Matter %	91.5	91.45	91.40	91.51	91.50
Calculated analysis					
Gross-Energy (Kcal/g)	417.71	414.98	414.84	414.66	414.48
Digestible-Energy (Kcal/g)	273.81	271.43	271.57	271.09	271.02
Metabolizable Energy (Kcal/g)	3202.52	3180.52	3180.56	3186.64	3185.70

FCe – Fermented *Canavalia ensiformis*

Growth performance of *Clarias gariepinus* fed FCe diets

Growth parameters measured, decreased as fermented *Canavalia ensiformis* meal inclusion levels increased. The highest and lowest mean weight gains (MWG) were recorded from the control and 100% FCe diets (6.78g/fish and 2.64g/fish) respectively (Table 3). There were significant differences (p<0.001) between the MWG of the diets. The relative growth rate (RGR) ranged from 162–674 %/fish. The highest value was from the control diet and the lowest value was from FCe 100% diet. There was a significant difference (p<0.001) among the diets. Specific growth rate (SGR) ranged from 0.46 %/day – 1.07%/day. The lowest value was recorded from FCe 75% diet and the highest was from the control diet. The highest survival was from control, FCe 25% and 50% diets (80.0%) and the lowest from 75% and 100% FCe diets (30.0% and 20.0%) respectively. There were significant differences (p<0.001) between the highest and lowest survival of fish-fed FCe diets. To compare the initial and final condition factors K1 and K2, the highest K1 was from fish fed 50% FCe diet (1.70) and the lowest was from fish fed 25% FCe diets (1.38) while the highest K2 was from fish fed the control diet (1.67) and the lowest was from fish fed FCe 50% diet (1.28). There was no significant difference (p>0.05) between K1 and K2.

Table 3: Growth Parameters and Survival Rate of *Clarias gariepinus* Fingerlings Fed Femented *Canavalia ensiformis* Diets

	Control 0%	FCe 25%	FCe 50%	FCe 75%	FCe 100%	SEM
TIW (g)	9.6±0.52	9.6±0.52	9.6±0.52	9.66±0.57	9.6±0.52	0.31^{ns}
TFW (g)	61.86±6.11^{a}	28.53±2.57^{b}	24.0±2.88^{b}	11.2±1.44^{c}	9.9±0.65^{c}	1.91***
MIW (g/fish)	0.96±0.05	0.93±0.05	0.96±0.05	0.96±0.05	0.96±0.05	0.03^{ns}
MFW (g/fish)	7.73±0.76^{a}	3.56±0.32^{b}	3.0±0.36^{bc}	2.8±0.36b^{bc}	2.5±0.20^{c}	0.25***
MWG (g/fish)	6.77±0.71^{a}	2.63±0.27^{b}	2.04±0.31^{bc}	1.84±0.31^{bc}	1.54±0.15^{c}	0.25***
MWWG (g/fish/week)	0.56±0.71^{a}	0.21±0.27^{b}	0.17±0.31^{bc}	0.15±0.31^{bc}	0.12±0.15^{c}	0.02***
MIL (cm/fish)	4.06±0.51	4.06±0.51	3.83±0.35	4.1±0.36	3.96±0.15	0.23^{ns}
MFL (cm/fish)	7.73±0.80^{a}	6.23±0.55^{b}	6.16±0.25^{b}	5.66±0.20^{b}	5.66±0.35^{b}	0.27***
RGR (%/fish)	673.0±0.71^{a}	282.7±0.27^{b}	212.5±0.31^{bc}	191.6±0.31^{bc}	160±0.15^{c}	34.43***
SGR (%/day)	1.06±0.71^{a}	0.69±0.27^{b}	0.57±0.31^{c}	0.45±0.31^{c}	0.48±0.15^{c}	0.03***
K1	1.43	1.38	1.7	1.39	1.54	0.25^{ns}
K2	1.67	1.47	1.28	1.46	1.5	0.21^{ns}
Survival (%)	80^{a}	80^{a}	80^{a}	30^{b}	20^{c}	2.12***

Mean ± S.D on the same row with different superscripts are significantly different (P<0.001), SEM-standard error of the mean, ***= (P<0.001), ^{ns} = not significant (p>0.05)

KEY: Total initial weight (TIW), Total final weight (TFW), Mean initial weight (MIW), Mean final weight (MFW), Mean weight gain (MWG), Mean weekly weight gain (MWWG), Mean initial length (MIL), Mean final length (MFL), Relative growth rate (RGR) and Specific growth rate (SGR).

FCe- Fermented *Canavalia ensiformis*

Feed intake and nutrient utilization of *Clarias* gariepinus fed fermented *Canavalia ensiformis (FCe)* diets

The highest mean feed intake (MFI) was obtained from fish fed a control diet (14.07g/fish) and the lowest was from fish fed FCe 100% diet (3.4g/fish), there was a significant difference (p<0.001) in the MFI of the fish fed the diets. The voluntary feed intake (VFI) ranged between 1.16 g/fish – 2.92g/fish. The highest value was from the fish fed the control diet and the lowest was from the fish-fed FCe 100% diet (Table 4). There was a significant difference (p<0.001) in the VFI of the fish fed the diets The highest feed acceptability index (FAI) was from the control diet (0.228%) and the lowest was from the fish fed FCe 100% diet (0.086%). There were significant differences (p<0.001) between the FAI of the control diet and FCe diets. The highest feed conversion ratio (FCR) was in the fish-fed FCe 50% diet (3.27) and the lowest was from the fish-fed control diet (2.07). There was a significant difference (P<0.001) between the FCR of the control diet and FCe diets. The protein intake ranged from 133.9–567.8 g/100 g diet/fish. The highest value was obtained from fish fed the control diet and the lowest was from fish fed FCe 100% diet.

Table 4: Feed Intake and Nutrient Utilization Indices of *Clarias gariepinus* Fingerlings Fed Fermented *Canavalia ensiformis* Diets

Parameters	Control 0%	FCe 25%	FCe 50%	FCe 75%	FCe 100%	SEM
TFI (g)	1181.88^{a}	642.6^{b}	561.12^{c}	338.52^{d}	285.6^{e}	2.19***
MFI (g/fish)	14.07±0.17^{a}	7.65±0.11^{b}	6.68±0.14^{c}	4.03±0.13^{d}	3.4±0.17^{e}	0.02***
BWFI (g/week)	196.98^{a}	107.1^{b}	93.52^{c}	56.42^{d}	47.6^{e}	0.31***
VFI (g/fish)	2.92±0.14^{a}	2.02±0.26^{a}	2.01±0.34^{a}	1.27±0.31^{b}	1.16±0.68^{b}	0.09***
FAI (%)	0.228±0.02^{a}	0.153±0.01^{b}	0.142±0.01^{c}	0.092±0.01^{d}	0.086±0.01^{e}	0.01***
FCR	2.07±0.23^{b}	2.9±0.40^{a}	3.27±0.45^{a}	2.19±0.41^{b}	2.2±1.13^{b}	0.00***
PI(g/100 g diet/fish)	567.8±0.01^{a}	307.9±0.01^{b}	268.2±0.01^{c}	158.9±0.01^{d}	133.9±0.01^{e}	1.03***
PER	0.13^{a}	0.052^{b}	0.042^{b}	0.038^{b}	0.036^{b}	0.00***

Mean ± S.D on the same row with different superscripts are significantly different (P<0.001)***, SEM- standard error of the mean

KEY: Total feed intake (TFI), Mean feed intake (MFI), Biweekly feed intake (BWFI), Voluntary feed intake (VFI), Feed acceptability index (FAI), Feed conversion ratio (FCR), Protein intake (PI), Protein efficiency ratio (PER),

FCe- Fermented *Canavalia ensiformis*

Mean Water Quality Parameters of Fish Fed Fermented *Canavalia ensiformis* Diets

The biweekly mean water quality parameters in the holding facilities during the feeding period are presented in figures (Figures 1-4). The highest temperature (27.8^{0}C) was recorded from 0% control and 25% FCe diets in week 2 while the lowest (23.1^{0}C) was from the 25% FCe diet in week 12. The highest dissolved oxygen (6.3 mg/l) was recorded from the 25% FCe diet in week 12 while the lowest (4.3mg/l) was from the 75% FCe diet in week 6. The highest pH (7.3) was recorded from the 75% FCe diet in week 4 while the lowest (4.8) from 100% FCe in week 10 and the highest ammonia (0.37mg/l) was from the 100% FCe diet in week 10 while the lowest (0.01mg/l) was from the five (0%, 25%, 50%, 75% and 100%) FCe diets in week 0. The results of the water quality parameters were significantly (p<0.001) different across the treatment groups.

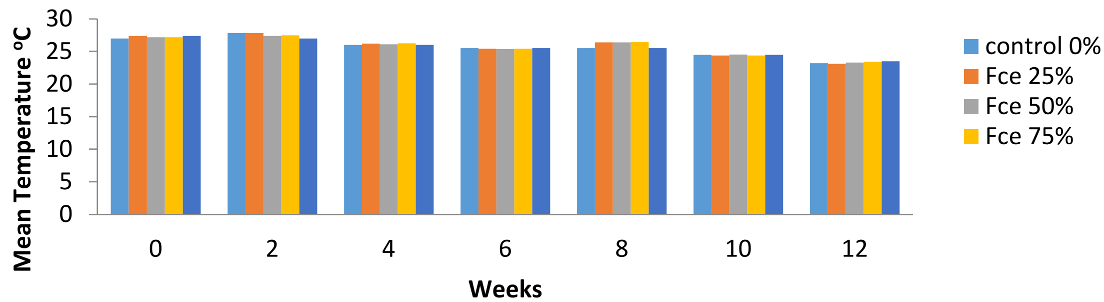

Figure 1: Biweekly Temperature (ºC) of Water Used to Raise *Clarias gariepinus* Fed FCe Diets

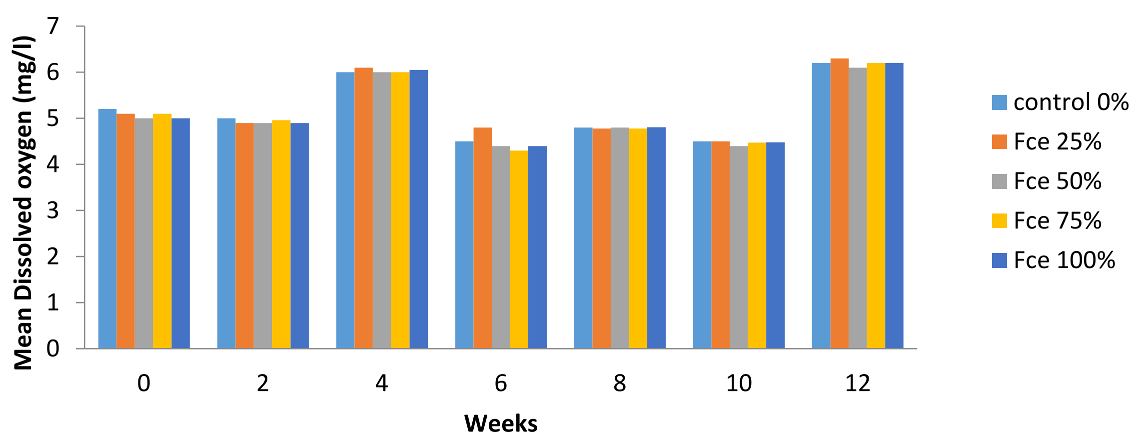

Figure 2: Biweekly Dissolved Oxygen (mg/L) of Water Used to Raise *Clarias gariepinus* Fed FCe Diets

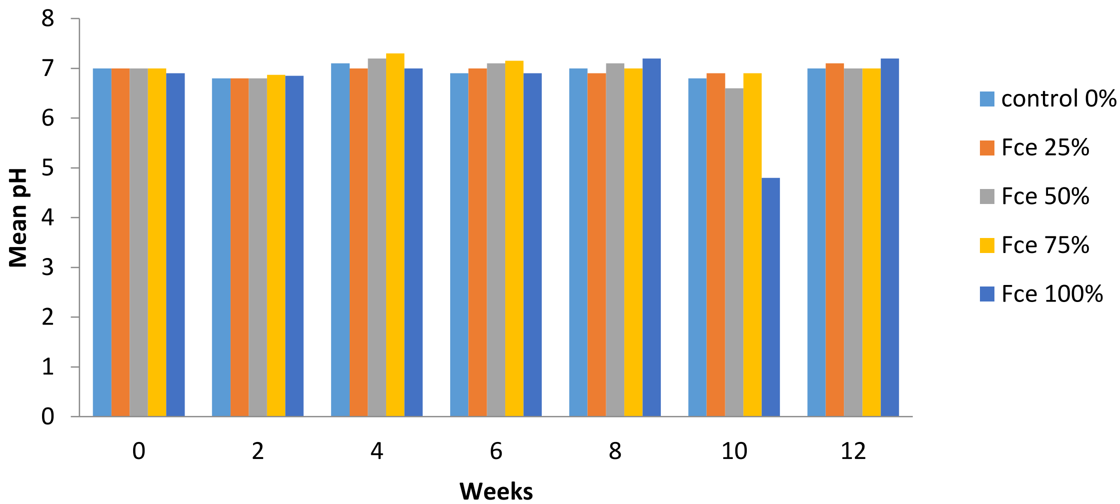

Figure 3: Biweekly pH of Water Used to Raise *Clarias gariepinus* Fed FCe Diets

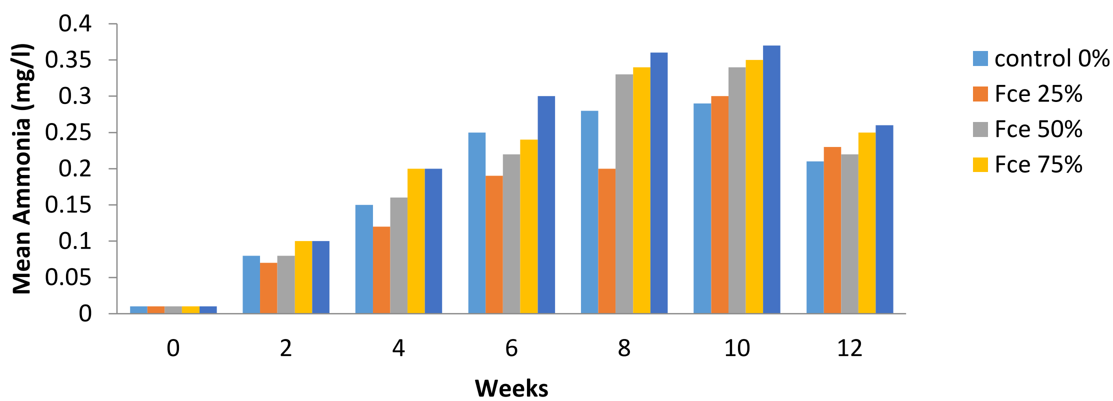

Figure 4: Biweekly Ammonia (mg/l) of Water Used to Raise *Clarias gariepinus* Fed FCe Diets

Nutrient digestibility coefficients of Clarias gariepinus fed FCe diets

Table (5) showed that the highest dry matter value was from the control diet (88.8%) and the lowest was from the 100% diet (82.3%) diet. The highest protein value was from the control diet (74.5%) and the lowest was from the 100% diet (57.8%). The highest lipid value was from the control diet (41.56%) and the lowest value was from the 75% diet (35.84%). The highest energy value was from the control diet (70.7%) and the lowest value was from the 100% diet (66.3%). There was a significant difference ($p<0.001$) in nutrient digestibility.

Table 5: Nutrient Digestibility Coefficients of *Clarias gariepinus* fed FCe Diets

Indices (%)	Control 0%	FCe 25%	FCe 50%	FCe 75%	FCe 100%	SEM
DMD	88.8±0.1[a]	87.1±0.1[b]	85.5±0.1[c]	83.9±0.1[d]	82.3±0.1[e]	0.05***
PD	74.5±0.1[a]	70.0±1.0[b]	65.0±1.0[c]	60.7±0.1[d]	57.8±0.1[e]	0.36***
LD	41.56±0.01[a]	40.66±0.00[b]	39.61±0.01[c]	35.84±0.00[e]	38.88±0.01[d]	0.01***
ED	70.7±0.1[a]	69.0±1.00[b]	68.0±1.00[bc]	67.1±0.1[cd]	66.3±0.1[d]	0.36***

Mean ± S.D on the same row with different superscripts are significantly different (P<0.001)*** , SEM- standard error of the mean

KEY: Dry matter digestibility (DMD), Protein digestibility (PD), Lipid digestibility (LD), Energy digestibility (ED)

FCe- Fermented *Canavalia ensiformis*

Basic Haematological indices of *Clarias gariepinus* fed FCe diets

Table (6) showed that the Highest PCV value was from the control diet (35.4%) and the lowest from the 100% diet (30.35%). The highest RBC value was from the control diet (1.40) and the lowest from 100% (1.14). The highest WBC value was from the 100% diet (22.94) and the lowest from the 50% diet (16.75). The Highest Hb value was from the control diet (10.6g/dl) and the lowest from the 100% diet (8.09g/dl). Highest MCV value was from the 100% diet (266.22dl) and lowest from 25% diet (246.73dl).

Table 6: Basic Haematological Indices of *Clarias gariepinus* Fed FCe Diets

Indices	Control 0%	FCe 25%	FCe 50%	FCe 75%	FCe 100%	SEM
PCV (%)	35.4±0.51[a]	34.05±0.81[b]	32.65±1.05[c]	31.46±0.47[cd]	30.35±0.36[d]	0.00***
RBC ($x10^6$)	1.40±0.41[bc]	1.38±0.39[c]	1.30±0.31[d]	1.25±0.26[ab]	1.14±0.15[a]	0.85***
WBC ($x10^3$)	20.5±1.25[bc]	19.35±1.02[c]	16.75±0.76[d]	21.83±0.84[a]	22.94±0.95[a]	0.01***
Hb (g/dl)	10.6±0.82[a]	9.85±1.3[ab]	9.23±0.44[abc]	8.75±0.76[bc]	8.09±0.85[c]	0.04**
MCV (dl)	252.85±1.08[b]	246.73±0.85[d]	251.15±0.16[c]	251.68±0.69[bc]	266.22±0.23[a]	0.01***
MCH (Pg)	75.71±0.83[a]	71.37±0.82[b]	71.0±1.00[b]	70.0±1.00[b]	70.96±0.97[b]	0.01***
MCHC (g/dl)	29.94±0.95[a]	28.92±1.03[ab]	28.26±0.81[abc]	27.81±0.82[bc]	26.65±1.05[c]	0.02**

Mean ± S.D on the same row with different superscripts are significantly different (P<0.01)** , (P<0.001) ***, SEM- standard error of the mean,

FCe- Fermented *Canavalia ensiformis*

KEY: Packed Cell Volume (PVC), Red Blood Cell (RBC), White Blood Cell (WBC), Haemoglobin (HB), Mean Corpuscular Volume (MCV), Mean Corpuscular Haemoglobin (MCH), Mean Corpuscular Haemoglobin Concentration (MCHC)

Economic evaluation of fermented *Canavalia ensiformis* (FCe) diets fed to *Clarias gariepinus*

The result of the production cost of six months FCe diets (Table 7) showed that the highest profit index was from FCe 75% diet (3.0) and the lowest was from the control and FCe 25% diets (2.8). The highest net profit was from the control diet (1142.5) and the lowest was from FCe 100% diet (838.6). The highest benefit-cost ratio was from FCe 25% diet (7.2) and the lowest was from the control and FCe 100% diets (6.5), there was a significant difference (p<0.001) in all the parameters across the treatment groups.

Table 7: Economic Evaluation of *Clarias gariepinus* Fingerlings Fed Fermented *Canavalia ensiformis* Diets for Six Months

Indices	Control 0%	FCe 25%	FCe 50%	FCe 75%	FCe 100%	SEM
COF ((₦)	479.5^{a}	443.5^{b}	407.5^{c}	363.5^{d}	336.5^{e}	0.18***
COFD (₦)	67.5^{a}	33.9^{b}	27.2^{c}	14.6^{d}	11.4^{e}	0.12***
EIC (₦)	207.5^{a}	173.9^{b}	167.2^{c}	154.6^{d}	151.4^{e}	0.18***
NPV (₦)	1350^{a}	1260^{b}	1170^{c}	1080^{d}	990^{e}	0.58***
PI (₦)	2.8^{b}	2.8^{b}	2.9^{ab}	3.0^{a}	2.9^{ab}	0.06***
IOC	120.5^{c}	161.4^{a}	160^{b}	97.3^{d}	95^{e}	0.36***
BCR	6.5^{c}	7.2^{a}	7.0^{b}	7.0^{b}	6.5^{c}	0.05***
NP (₦)	1142.5^{a}	1086.1^{b}	1002.8^{c}	925.4^{d}	838.6^{e}	0.05***

Values on the same row with different superscripts are significantly different (P<0.001)*** , SEM- standard error of the mean,
FCe- Fermented *Canavalia ensiformis*
KEY: Cost of feed (COF), Cost of feeding (COFD), Estimated investment cost (EIC), Net profit value (NPV), Profit index (PI), Incidence of cost (IOC), Benefit cost ratio (BCR), Net profit (NP)

DISCUSSION

In applied research and practice, weight gain and specific growth rate are usually considered the most important measurement of the productivity of diet (Agbo *et al*., 2015; Oyetunji *et al*., 2021) and a reliable indicator of economic evaluation as a marketable product. The fact that growth was recorded from all experimental diets indicated that the fish was able to convert the protein in the feed to muscles as reported by Sogbesan *et al*. (2008). The control treatment had higher values than the FCe treatment, in terms of FCe treatments, better performances were recorded from 25% and followed by 50% FCe treatment which agrees with the report of Agbo, *et al*., 2015 on fermented Sunflower meal diet (Hasanna *et al*., 2018).

This study disagrees with Olatunde (2003) who reported better performance in (D3) fish fed 20% fermented sicklepod seed meal diet. In this study, the reason for the difference in acceptability of the diets among treatments agrees with earlier reports by authors (Bake, *et al*., 2015; Riche, *et al*., 2001; Riche, *et al*., 2004), suggested that when alternative protein sources especially plant protein sources are used in the fish diet, palatability and attractiveness of the diets are usually affected as seen in the case of treatment 3-5 (50, 75 and 100%) inclusion of fermented C. ensiformis diets. As suggested by Ahmad (2008) proper utilization of dietary protein is dependent on the good quality or amino acid balance of the protein sources. Nguyen *et al*. (2009) stated that the weight gain (WG) of young fish is a reliable indicator of nutritional efficiency. This present study showed that fermentation and inclusion levels both affected the weight gained. This agrees with the findings of previous studies (Nguyen *et al*., 2009; Diarra, 2021; Rodriguez-Serna, *et al*., 1996; Jimoh, *et al*., 2014 and Francis, *et al*., 2001) that one of the major limitations of using alternative plant protein sources in fish feeding is a decrease in palatability and the presence of anti-nutrition factors in the ingredient. The reduction in feed intake due to the unpalatability may lead to deterioration in growth performance and feed utilization (Riche *et al.,* 2004) A reduction in growth and nutrient utilization in fish fed 75% and 100% FCe diets was observed; this reduction observed could be attributed not only to the dietary amino acid profile of the ingredient but also to the presence of anti-nutritional factors. This assertion is in line with the findings of (Siddhuraju *et al*., 2003; Davies, *et al*., 2000; Jimoh *et al*., 2011) that higher inclusion levels of most plant protein source base meals resulted in poor growth and nutrient utilization. The best feed conversion ratio (FCR) was recorded in the control diet, followed by the 25% FCe diet which is an indication of an optimum level of utilization of the diet by the *Clarias gariepinus* fingerlings, this corresponds with Wang *et al*. (2021) who stated that the lower the FCR, the better the feed utilization by the fish and observations made by (Bake *et al*., 2020; Balogun *et al*., 2016; Shabbir *et al*., 2003) in related studies on feeding trials. The protein efficiency ratio (PER) is known to be regulated by the non-protein energy input of the diet and is a good measure of the protein-sparing effect of lipids and/or carbohydrates (Jabeen *et al*., 2004; Ochang *et al*., 2007). The general well-being of the fish-fed fermented *Canavalia ensiformis* diets is expressed by the condition factor (K). The values were above 1.0 and agreed with Tibbets *et al*. (2005). Survival rate was higher in control, 25% and 50% treatments while lower in 75% and 100% treatments and this might be due to the presence of antinutrients in the diets or other extraneous factors which agreed with Jamabo *et al*. (2020) who reported that mortality might not be due to the antinutrients in the diets alone but also to any other extraneous factors such as stress resulting from handling.

These water quality parameters ranges for all the treatment diets were within the tolerable limit in aquaculture (Tibbets, *et al.,* 2005; Wei, *et al*., 2019; Adekoya, *et al*., 2004; Omotayo, *et al*., 2011) recommended Dissolved Oxygen levels of between 4-8mg/litre in the pond and the values observed during the experimental period fall within this range. Lawson, 1995 reported that when the DO level is consistently between 1.5-5 mg/litre, fish will be alive, but feed intake will reduce. The growth rate will also reduce and high Feed Conversion Ratios (FCR) will be recorded. When DO levels are lower than 1.5mg/litre, fish will be stressed and they will die (Lawson, 1995). The periods of achieving desired weights in fish will be lengthened and ultimate loss on investment will occur. The range in the average temperature recorded during the experimental period was probably because all the treatments were outdoors and faced with environmental factor variations. Anjulo *et al.* (2021) reported that for African Catfish, an acceptable water temperature range is between 26ºC to 32ºC. When the water temperature in the ponds consistently stays between 16ºC and 26ºC, feed intake reduces and the fish growth rate also drags tremendously. A farmer recorded high FCR, and the fish stressed. Prolonged stress can open up the fish to opportunistic infections. When fish are consistently exposed to temperatures below 15ºC, fish growth ultimately stops with eventual mortality. Low temperature negatively affects the rates at which wastes are converted into water. However, when the water temperature is above 32ºC, the resultant effect on the African Catfish is not good at all (Anjulo *et al*., 2021), this is because Oxygen is not readily soluble in very warm water. High temperatures in ponds will stress the fish and eventually lead to death (Anjulo *et al.*, 2021). Anjulo *et al*. (2021) reported that pH is the level of the Hydrogen ion present in the water. For the fish in the pond, the acceptable pH value is between 6.5 - 7.5. Fish will die when pH falls below 4 owing to the water's acidity. Fish will still be alive when the pH is continuously between 4 and 6, but they will grow slowly because of stress. The amount of food consumed will be drastically reduced. Also, FCR will be very high. Low pH in pond water is a sign of excessive CO2 (carbon dioxide) levels, according to the astute fish farmer. Fish growth will also be slowed down by pond water with high pH levels of 9 to 11. Eventually, fish will die when pH values exceed 11. Ionized ammonia, which is less hazardous to fish, is produced in greater quantities when the pH is low. High pH levels in water have the opposite effect (Anjulo *et al*., 2021). The toxic levels for unionized ammonia for short-term exposure usually lie between 0.6 and 2.0 mg/l for pond fish, and sub-lethal effects may occur at 0.1 to 0.3 mg/l which corresponds to the findings in this study. The effects of high concentrations of ammonia on fish include damage to tissue, gill and kidney, reduction in growth, malfunctioning of the central nervous system, reduction in the oxygen-carrying capacity of the haemoglobin, poor feed conversion, poor growth, reduced disease resistance, susceptibility to infections, decrease fecundity, reduction of population sizes and fish kill. The manifestation of these effects depends on the life stage of the fish, the period of exposure and the tolerance of the species (Mustapha *et al*. 2016).

There is a slight increase in the digestibility coefficient of fishes fed fermented *Canavalia ensiformis* (FCe) at a 25% level to those fed toasted *Canavalia ensiformis* (TCe) diets, which agrees with the report of Fagbenro *et al.* (2007) that the likely thermostable antinutrients in the jack bean seeds formed a good proportion of the solubilized and removed nitrogenous compounds, which might be partly responsible for the improvement in the nutritive values of processed jack bean seeds. The high dry matter digestibility by *Clarias gariepinus* may be due to the crude fibre content of the diets. This finding is similar to the report (Ologbobo, 2006) that dry matter digestibility could be affected by the fibre content of the diet. The reduction in crude protein digestibility may be due to the presence of the anti-nutrients. Tannins have been implicated in reducing protein digestibility (Koprucu *et al*., 2005). The relatively high apparent digestibility coefficient of energy recorded in this study is similar to the values recorded by (Jimoh, *et al*., 2010; Adeparusi *et al*., 2002) of the same seed meal fed to *O. niloticus*. Osuigwe *et al.* (2005) and Bhagya *et al.* (2010) revealed that cooking improved gross energy digestibility.

The decrease in haemoglobin, haematocrit, and RBC observed with increasing dietary fermented Canavalia seed meal as compared with the control diet could be a result of the anti-nutritional factors still present in FCe diets at higher inclusion levels which agrees with the report of Gboshe *et al.* (2020). Some of these anti-nutritional factors are known to cause some negative effects on some basic haematological parameters. Con-A causes agglutination of red blood cells in monogastric (Ogunji *et al*., 2008), while saponins are known to cause erythrocyte haemolysis and reduction of blood (Garcia-Abiado *et al*., 2004). Probably the increasing presence of anti-nutritional factors in increasing dietary Canavalia caused the inferior haematological parameters observed in *C. gariepinus* fed such diets. This agrees with the findings of Jimoh *et al*. (2015) that nutritional toxicity is associated with anaemia. Adogu *et al*. (2015) equally observed that gossypol an anti-nutritional factor found in some legumes severely reduced blood PCV and Hb concentration in catfish and tilapia with adaptation to an ambient water temperature range of 23-28oC. When viewed from the perspective of diet processing type, it was observed that *C. gariepinus* fed the control diet had PCV, RBC count, WBC count and Hb concentration that were higher and significantly ($p<0.001$) different from the values of those fed fermented Canavalia diets which were, in turn, higher than those fed toasted Canavalia diets. According to Seena *et al.* (2006), RBC greater than 1x106/mm3 is considered high and is indicative of the high oxygen-carrying capacity of the blood which is characteristic of fishes capable of aerial respiration and with high activity. The better performance of *C gariepinus* fed fermented Canavalia diets to those fed toasted Canavalia diets is an indication that fermentation significantly improved the quality of FCe meals. The improvement may be due to among other factors inactivation of the anti-nutritional factors present in Canavalia as earlier reported by Megbowon *et al*. (2014) and the transformation of some of the component nutrients to non-toxic more readily digestible absorbable forms Opiyo *et al.* (2013). It is importance to note that despite the reduction in the levels of haematological values observed, they were still within the normal ranges reported for *C. gariepinus* (Ezewudo *et al*., 2015).

The economic evaluation of feeding *Clarias gariepinus* fingerlings with the experimental diets showed that FCe 25% had the highest benefit: cost ratio. The positive benefit: cost ratio recorded in all the diets, showed that *Clarias* fingerlings can be economically reared on all diets. However, the result further revealed that the substitution of fermented *Canavalia ensiformis* seed meal for soybean meal lowered the cost of diet production, which is an indication of a more cost-efficient and cheaper non-convectional ingredient relative to the soybean meal by Opiyo *et al.* (2013) and Sogbesan *et al.* (2008). This is similar to the report that non-conventional feed resources (NCFRs) are very cheap by-products or wastes from agriculture, farm-made feeds and processing industries (Sogbesan *et al.*, 2008). It implies that fermented *Canavalia ensiformis* seed diets at a 25% inclusion level can be fed to *Clarias gariepinus* fingerlings with better growth performance, haematology, digestibility and economic evaluations.

CONCLUSION

Optimal growth and weight gain in this study were obtained at an inclusion level of 25% of fermented *Canavalia ensiformis* meal. It is concluded that fermented *Canavalia ensiformis* meal can be used as a growth-promoting agent in the diet of *Clarias gariepinus* with better haematology and economic evaluation.

This study discovered that Fermented *Canavalia ensiformis* diets can be beneficial for *Clarias gariepinus* fingerling's growth. This study will help the researchers to uncover the critical areas of inclusion levels that many researchers were not able to explore.

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