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*Full Length Research Paper*

# Formulating efficient and affordable feeds for *Clarias gariepinus* (Burchell, 1822) based on locally available ingredients in Benin, West Africa

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**An affordable feed for the growth of African catfish (*Clarias gariepinus*) juveniles was developed in Benin as an alternative to high-cost imported feed. Initially, locally available fish feed ingredients were inventoried. Subsequently, six feed formulas (Feeds 1 to 6) were developed using linear algebra software, and the feeds were pelleted using a locally built extruder. The protein content of the formulated feeds ranged from 366 to 405 g kg$^{-1}$, fat content from 84 to 111 g kg$^{-1}$, and carbohydrate content from 239 to 330 g kg$^{-1}$. Among the formulations, Feed 1 (protein: 390 g kg$^{-1}$, fat: 84 g kg$^{-1}$, carbohydrates: 330 g kg$^{-1}$) closely matched the nutritional composition of the imported feed (protein: 424 g kg$^{-1}$, fat: 84 g kg$^{-1}$, carbohydrates: 346 g kg$^{-1}$) commonly used by most fish farmers in Benin for catfish. Additionally, it had the lowest ingredient cost (0.46 US$ kg$^{-1}$). Following this, the growth of *C. gariepinus* fingerlings was evaluated when fed with Feed 1 in a 56-day exploratory trial. The weight gain, feed intake, and protein intake of fish fed with Feed 1 were significantly lower (p = 0.000 for each parameter) than those fed with the control feed. However, Feed 1 exhibited a good Feed Conversion Ratio (1.25) and an acceptable protein efficiency ratio (2.05).**

**Key words:** Extrusion, feed formulation, feeding trial, feed ingredients, nutritional composition.

## INTRODUCTION

Fish farmers require nutritious and affordable feeds to ensure the profitability of their businesses. Babalola (2010) highlighted that fish feed alone constitutes approximately 75% of the total costs of fish production in West African countries. According to Adéyèmi et al. (2020), fish farming in Benin relies entirely on imported feeds that are high in cost. The primary determinants of this market price for fish feed include the protein content

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and its quality (Hamre et al., 2013; Shepherd and Jackson, 2013). Therefore, protein emerges as the fundamental macronutrient that must be prioritized in the selection of ingredients for feed formulation. This necessity propels researchers globally to explore alternative, sustainable protein sources that offer comparable nutritional value to fishmeal, the primary component of commercial fish feed. Locally available ingredients for fish feed formulation are preferred due to their lower transportation costs. In Benin, as well as in many other West African countries, fish feed formulation is still in its early stages of development. Most local attempts to create suitable fish feed have failed due to the lack of information and expertise. Presently, fish farming heavily relies on imported feed despite the availability of potential ingredients for local feed formulation.

The utilization of agricultural by-products, such as soybean meal (Imorou Toko, 2007), cottonseed meal (Monentcham et al., 2010; Ashiru et al., 2015), and groundnut cake meal (Tiamiyu et al., 2013), in fish feed formulation has yielded promising outcomes in various countries. Furthermore, there has been a reported option to substitute fishmeal with alternative animal protein feedstuffs. For instance, the incorporation of garden snails - *Limicolaria aurora* - (Sogbesan et al., 2006), tadpoles - *Bufo maculate* - (Sogbesan and Ugwumba, 2007), termites - *Macrotermes subhyalinus* - (Sogbesan and Ugwumba, 2008), maggots (Wang et al., 2017), earthworms (Djissou et al., 2017), and black soldier fly larvae (Belghit et al., 2019; Agbohessou et al., 2021) in fish feed formulation has been documented. Additionally, certain animal by-product meals, such as poultry by-product meal, hydrolyzed feather meal, bone meal, and blood meal, have been noted to possess high protein levels and essential amino acid (EAA) profiles conducive for fish feed formulation (Shapawi et al., 2007). Ingredients sourced from plants, such as *Leucaena* leaves and Moringa oleifera leaf meals, have also been incorporated into feed formulations for *Clarias gariepinus* in Nigeria and Benin (Chabi et al., 2015; Tiamiyu et al., 2015).

These authors achieved a protein content of 400 g kg$^{-1}$ in feed formulations for *C. gariepinus*. According to Adéyèmi et al. (2020), the African catfish, *C. gariepinus*, is one of the most farmed fish in Benin (West Africa), despite its high protein requirement (up to 420 g kg$^{-1}$ for catfish juveniles). The fish is often smoked and widely used in sauces (Amisah et al., 2009). It is one of the most important fish species in commercial aquaculture due to its high fecundity and the quality of its meat, which is highly appreciated by consumers (Amisah et al., 2009). The production of African catfish significantly contributes to food security and income generation for thousands of West Africans.

These benefits can be further enhanced by making low-cost and nutritionally efficient feeds available to fish farmers. Consequently, this study was conducted to rank and identify promising feed ingredients available in Benin and to formulate low-cost feeds that meet the nutrient requirements of African catfish.

## MATERIALS AND METHODS

### Period and area of study

The study was conducted from September 2019 to February 2020 in Benin, West Africa, located at coordinates 9°30'N 2°15'E.

### Ranking fish feed ingredients according to feed value

Fish feed ingredients from Benin were previously characterized by Adéyèmi et al. (2020). To rank the collected feed ingredients, their feed value was calculated using the formula outlined in Table 1, which considers the feed nutritional index (Ni), the availability index (Avi), and the cost index (Ci). While other nutritional properties such as amino acid and fatty acid composition, as well as ingredient digestibility, are important factors to consider when calculating feed value, budget constraints prevented their inclusion in this study. However, the calculated feed value index proved to be robust enough to effectively rank the feed ingredients.

### Formulation and production of feeds based on available local feed ingredients

Based on the nutritional composition of promising local feed ingredients and following the methodology outlined by Chakeredza et al. (2008), six nutritionally balanced and low-cost feeds for catfish fingerlings (Feed 1, Feed 2, Feed 3, Feed 4, Feed 5, Feed 6) were formulated. Fishmeal imported from Senegal, soybean meal, cottonseed meal, premix, and methionine were procured from the livestock feed center "La Confiance" in Abomey-Calavi, Benin. Trash fish, cassava chips, and red palm oil were purchased from local markets in Abomey-Calavi. Whole garden snail and poultry viscera were obtained from open local markets in Sakété and Cotonou, Benin, respectively. Brewer's yeast slurry was sourced from SOBEBRA, a beer brewing company in Cotonou. Each dry ingredient was ground in a hammer mill with a screen size of 1 mm. Subsequently, predetermined quantities of ground ingredients were mixed to obtain 3 kg of each formulation. Brewer's yeast slurry and/or water were added to achieve a moisture content of 300 g kg$^{-1}$. Feeds were mixed with palm oil and extruded using a locally constructed low-cost extruder fitted with a die with 4 holes of 6.0 mm diameter each. The extrudates were dried in a conventional batch hot-air dryer at 60ºC for 3 h. Each formula was produced in duplicate. A frequently used imported feed in Benin was utilized for comparison purposes.

### Nutritional characterization of feeds

The nutrient content of feeds was evaluated according to AOAC (2005) methods. Dry matter content was determined by oven drying at 105 °C for 48 h. Crude protein was determined using the micro-Kjeldahl method (Kirk, 1950). The amount of crude protein was calculated by multiplying the percentage of nitrogen in the digest by 6.25 (Mariotti et al., 2008).

Crude lipid was determined by Soxhlet extraction using petroleum ether as a solvent. Ash content was obtained by muffle

**Table 1.** Calculation of the feed value of potential feed ingredients.

| Parameter | | Requirement for juveniles of *C. gariepinus* (g $kg^{-1}$ of feed) | Index calculation |
|---|---|---|---|
| Nutritional index | Protein index (Proti) | 400 - 430[1] | $\frac{\text{Protein content}}{41.5}$ |
| | Lipid index (Fi) | 87 - 170[1] | $\frac{\text{Lipid content}}{12.85}$ |
| | Fiber index (Fbi) | 31 - 37.1[2] | $\frac{3.41}{\text{Fiber content}}$ |
| | Ash index (Ai) | 68 - 80[2] | $\frac{\text{Ash content}}{7.4}$ |
| | Carbohydrate index (Carbi) | 150 - 380[3] | $\frac{\text{Carbohydrate content}}{26.5}$ |
| | Nutritional index (Ni) | **Ni =** 4×Proti + Fi + Fbi/2 + Ai + Carbi | |
| Availability index (Avi) | | Average availability (ton/month) cited by respondents | |
| Cost index (Ci) | | Average (US $/kg) of cost cited by respondents | |
| Feed value = 2Ni + Avi + 1/Ci | | | |

[1]Ali and Jauncey, (2005), [2]Based on label information on bags of imported feed, [3]Ali and Jauncey (2004)
Source: Our Data (2020).

furnace at 550°C, while the carbohydrate content was calculated by difference: 100% - (% moisture + % protein + % lipids + % ash). Gross energy was calculated according to Alegbeleye et al. (2012) based on an estimated 23.65 kJ $g^{-1}$ for protein, 40 kJ $g^{-1}$ for fat and 16.8 kJ $g^{-1}$ for carbohydrate. The measurements of each studied parameter were conducted in duplicate for each sample of the two different productions, resulting in a total of 4 repetitions. Relevant information about amino acid and fatty acid composition of ingredients was collected from various literature sources (National Research Council, 1993; Robinson et al., 2001; Craig et al., 2017).

### Fish feeding trial

An exploratory study was conducted to examine the overall behavior of a new fish feed formula whose nutrient composition closely resembled that of the commonly used imported commercial feed. This rapid observational study took place at the Food Africa farm in Porto-Novo (Latitude: 6° 29', Longitude: 2° 36'), Benin. Three hundred and sixty (360) Clarias gariepinus fingerlings were obtained from a reputable fish farm (TONON Foundation) in Abomey-Calavi (Latitude: 6° 26', Longitude: 2° 21'). They were transported in an open container to the experimental site and acclimatized for one week, during which they were fed with the control feed containing approximately 420 g kg-1 crude protein. The average initial body weight of the fish was approximately 5 g.

For this exploratory study, two identical concrete tanks (5 x 5 x 1.5 m) were utilized: one containing fish fed with the formulated feed and the other with fish fed with the control feed. Each concrete tank was divided into three compartments (1.5 x 3 x 1.5 m) for three repetitions, resulting in a total of six experimental treatment units. Prior to distribution, the fingerlings were starved overnight, weighed (approximately 7.3 g), and then randomly released at a rate of sixty (60) fish per experimental treatment unit. The tanks were supplied with borehole water without any additional treatment, and the fish were fed ad-libitum three times daily for 56 days. Feeding response and water quality were monitored. pH, dissolved oxygen (DO), and water temperature were measured using a pH meter (WTW pH 3210) and a DO meter (HANNA HI 9146), respectively, twice a week before morning feeding. The water in the tanks was changed every two weeks during the experiment. Daily feed intake was measured per tank, and batch weighing of fish was conducted every two weeks. Growth performance and nutrient utilization of the fish were evaluated according to Castel and Tiews (1980) based on the following parameters:

1) Initial Mean Weight (g $fish^{-1}$) = Initial biomass (Wi) / Initial number of fish
2) Final Mean Weight (g $fish^{-1}$) = Final biomass (Wf) / Final number of fish
3) Weight gain per day (g $fish^{-1}$) = (Final Mean Weight - Initial Mean Weight) / days of feeding.
4) Specific growth rate (%/fish/day) = [(log Final biomass – log Initial biomass) / days of feeding] × 100.
5) Protein intake (g) = Feed intake × Feed protein content /100.
6) Feed Conversion Ratio = Feed intake (g) / Fish weight gain (g)
7) Protein Efficiency Ratio = Fish weight gain (g) / Protein intake (g).

### Statistical analysis

Descriptive statistics were followed by a one-way Analysis of Variance (ANOVA) to evaluate significant differences among means. When significant differences ($p < 0.05$) were observed, data were separated using Tukey's post-hoc test. Principal Component Analysis (PCA) was performed to cluster feeds based on their nutritional characteristics, including protein, fat, ash, carbohydrate contents, and gross energy. Statistical analysis was conducted using Minitab 18 Computer Software (Minitab LLC, Pennsylvania State University).

## RESULTS AND DISCUSSION

### Ranking fish feed ingredients according to feed value

Feeds for *C. gariepinus* typically incorporate a variety of ingredients to meet the nutritional requirements of the fish. A balanced feed for optimal growth rate and feed

**Table 2.** Local fish feed ingredients of Benin ranked according to their calculated feed value.

| Ingredients | Ni | Avi | Ci | Feed value (2Ni + Avi + 1/Ci) |
|---|---|---|---|---|
| ***Protein-rich ingredients of animal and microbial origin*** | | | | |
| Dried brewer's yeast[4] | 15.94 | 4 | 0.09 | 47.00 |
| Trash fish | 18.14 | 2 | 1.12 | 39.27 |
| Garden snail | 16.96 | 1 | 1.29 | 35.78 |
| Fish meal (low quality) | 10.96 | 8 | 0.52 | 31.85 |
| Poultry viscera | 11.84 | 0.1 | 0.37 | 26.48 |
| Fish meal (high quality) | 12.99 | 0.1 | 2.59 | 26.48 |
| ***Protein-rich ingredients of plant origin (grain by-products)*** | | | | |
| Cottonseed meal | 6.66 | 11 | 0.33 | 27.34 |
| Soybean meal | 6.93 | 6 | 0.67 | 21.36 |
| Soybean | 6.99 | 5 | 0.52 | 20.94 |
| Brewer's spent grain (industrial) | 5.88 | 1 | 0.17 | 18.65 |
| Rice bran | 5.52 | 2 | 0.18 | 18.60 |
| Brewer's spent grain (local beer) | 6.70 | 0.1 | 0.20 | 18.50 |
| Maize bran (Mawè waste product) | 4.42 | 0.1 | 0.13 | 17.85 |
| Wheat bran | 5.13 | 3 | 0.27 | 16.96 |
| Maize bran (industrial) | 5.03 | 3 | 0.29 | 16.45 |
| Soybean bran (Soya cheese waste product) | 5.96 | 0.1 | 0.36 | 14.81 |
| ***Protein-rich ingredients of plant origin (leafy vegetables)*** | | | | |
| Moringa leaf | 6.43 | 1 | 0.52 | 15.78 |
| Kapok-tree leaf | 5.91 | 0.1 | 0.34 | 14.86 |
| Azolla leaf | 6.07 | 0.1 | 0.52 | 14.16 |
| ***Starch-rich ingredients*** | | | | |
| Cassava chips | 4.45 | 10 | 0.43 | 21.23 |
| Maize | 4.64 | 9 | 0.37 | 20.99 |
| Lafun | 4.25 | 3 | 0.34 | 14.44 |
| Tapioca | 4.50 | 3 | 0.52 | 13.92 |
| ***Other fish feed ingredients*** | | | | |
| Oyster shell | 13.91 | 109 | 0.26 | 140.66 |
| Palm kernel cake | 5.66 | 10 | 0.28 | 24.89 |

Ni = Nutritional index; Avi = Availability index; Ci = Cost index, [4]- Obtained by drying brewer's yeast slurry at Laboratory. Fishmeal (Low quality, imported from Senegal), soybean meal, cottonseed meal, brewer's spent grain (industrial), rice bran, wheat bran, maize bran (industrial), oyster shell and palm kernel cake were purchased at livestock feed centre "La Confiance" at Abomey-Calavi/Benin, Fishmeal (high quality, imported from U.K.) was collected at local fish feed producer in Zè/Benin, Trash fish, cassava chips, maize, soybean were purchased at local markets in Abomey-Calavi/Benin, Garden snail was purchased at open local markets in Sakété, Benin, Poultry viscera, lafun and tapioca were purchased at open local markets in Cotonou, Benin, Brewer's yeast slurry was obtained from SOBEBRA, a beer brewing company in Cotonou, Benin, Brewer's spent grain (of local beer), maize bran (mawè by-product), soybean bran (soya cheese by-product) were collected at the local producers of Tchoukoutou, of Mawè and of Soya cheese, respectively at Abomey-Calavi, Benin, Moringa leaf was collected at the farm of the Faculty of Agricultural Science of the University of Abomey-Calavi, Benin, Azolla and Kapok-tree leaves were collected at the local fish producer in Tori-Bossito, Benin.
Source: Our Data (2020).

conversion efficiency for catfish should ideally contain 380-425 g $kg^{-1}$ protein, 100-110 g $kg^{-1}$ fat (Jimoh et al., 2013; Djissou et al., 2016), and 150-380 g $kg^{-1}$ carbohydrates (Ali and Jauncey, 2004). Beninese fish feed ingredients were ranked based on their feed value, as shown in Table 2. Adhering to these indices is crucial, especially from the perspective of potential industrial exploitation of the proposed formulas. Fish feed

**Table 3.** Chemical composition (g kg$^{-1}$ of Dry Matter) and Cost (US$ kg$^{-1}$) of the ingredients.

| Raw materials | DM | Ash | Proteins | lipids | Fibers | Carbohydrates | Cost |
|---|---|---|---|---|---|---|---|
| Fishmeal (low quality) | 890.0 | 375.2 | 410.8 | 78.0 | 19.4 | 26.0 | 0.52 |
| Trash fish | 945.0 | 242.7 | 679.0 | 78.8 | 2.2 | 00.0 | 1.12 |
| Brewer's yeast slurry | 125.0 | 12.0 | 64.0 | 4.0 | 3.0 | 45.0 | 0.09 |
| Dried brewer's yeast | 947.0 | 29.9 | 422.5 | 16.1 | 52.4 | 478.5 | 1.64 |
| Whole garden snail meal | 970.6 | 698.0 | 171.3 | 37.3 | 3.2 | 64.0 | 1.29 |
| Poultry viscera meal | 837.3 | 66.4 | 586.0 | 281.5 | 5.5 | 00.0 | 0.37 |
| Cottonseed meal | 880.0 | 66.7 | 436.7 | 14.9 | 120.0 | 361.7 | 0.33 |
| Soybean meal | 890.0 | 60.3 | 453.0 | 30.7 | 118.8 | 346.0 | 0.67 |
| Moringa leaf meal | 905.4 | 83.3 | 347.2 | 44.6 | 163.5 | 430.3 | 3.45 |
| Cassava chips flour | 860.0 | 35.3 | 16.4 | 6.5 | 30.9 | 801.8 | 0.43 |

Source: Adéyèmi et al. (2020).

formulation approaches that integrate such parameters with relevant local feed characteristics are rare. Hence, our feed value index of ingredients considers their nutrient content as well as their accessibility in terms of quantity and cost.

The ranking test identified brewer's yeast slurry, trash fish meal, fishmeal, poultry viscera, garden snail meal, cottonseed meal, and soybean meal as promising ingredients in the category of protein-rich ingredients for fish feed formulation. Whole garden snail meals and fishmeal (low quality) are also significant sources of minerals. Cassava chips stood out as starch sources. Moringa leaf meal exhibited a relatively low feed value due to its poor market availability. However, this suggests that Moringa can become an interesting fish feed ingredient if its availability is improved. Currently, several projects are underway in the country to promote the cultivation of Moringa. Interestingly, Moringa leaf powder can also significantly contribute to the micronutrient content of feeds (Agbogidi and Ilondu, 2012). The proximate composition and costs of these promising ingredients are presented in Table 3 (Adéyèmi et al., 2020).

Indeed, trash fish consists of the residue of smoked and dried small fish, comprising a mixture of heads, skeletons, viscera, scales, and whole fish, commonly sold in most open African markets. Sotolu (2009) reported that fish waste meal or trash fish meal is capable of supplying adequate nutrients in a manner comparable to high-quality fishmeal, containing as much as 788 g kg-1 DM of protein. The author noted that incorporating 150 g kg$^{-1}$ trash fish in feed for *C. gariepinus* to replace conventional fishmeal resulted in similar growth performances. Fishmeal sold in Beninese markets is of low and variable quality, with crude protein contents ranging from 196 to 411 g kg$^{-1}$ DM (Adéyèmi et al., 2020).

Another by-product discussed is brewer's yeast slurry, obtained from the brewing industry, which has been shown to positively influence immune responses and the growth of some fish species (Oliva-Teles and Goncalves, 2001) and enhance the intestinal health of fish (Zhou et al., 2018). Garden snails and poultry viscera, although potential animal protein sources for fish feeds, are yet uncommon. Sogbesan et al. (2006) tested the possibility of using garden snail (*Limicolaria aurora*) meat meal as a protein source in catfish feeds and recommended its incorporation at 250 g kg$^{-1}$ for optimum fish growth and nutrient utilization. Whole garden snail is also of interest because its shell is a valuable source of minerals. Bhaskar et al. (2015) recommended a maximum of 250 g kg$^{-1}$ of poultry viscera meal to replace fishmeal in feeds for Koi fish (*Anabas testudineus*).

Soybean meal (Glycine max) is widely used in animal feed, especially in aquaculture, due to its availability, price, and good nutritional value. It is considered the best plant protein source for fish feeds, with a high protein content and a favorable amino acid profile (Zhou et al., 2005), and can largely replace fishmeal in African catfish feeds (Goda et al., 2007). However, plant protein is deficient in essential amino acids such as methionine and lysine, which can be overcome by adding one or more essential amino acid supplements. According to Fagbenro and Davies (2001), methionine is the most limiting amino acid in soybean protein.

Cottonseed meal, a by-product of cottonseed oil extraction, has been used in feeds for aquatic animals for many years (Imorou Toko, 2007; Yuan et al., 2019). Among the protein sources of plant origin, cottonseed meal has been considered an alternative for fishmeal in fish feed because of its high protein content, relatively low price, and sufficient availability.

Trypsin inhibitors are present in cottonseed meal, but these can be destroyed by heat treatment during processing (Elmaki et al., 2007). Likewise, free gossypol, a natural toxin in cottonseed meal, can be reduced by extrusion; however, stable conjugated gossypol can

**Table 4.** Formulated feed composition (g $kg^{-1}$ wet basis)[5].

| Ingredients | Feed 1 | Feed 2 | Feed 3 | Feed 4 | Feed 5 | Feed 6 |
|---|---|---|---|---|---|---|
| Fishmeal (low quality) | 0 | 120 | 120 | 0 | 0 | 97 |
| Trash fish | 150 | 150 | 150 | 270 | 270 | 147 |
| Garden snail meal | 0 | 0 | 0 | 0 | 0 | 10 |
| Poultry viscera meal | 0 | 0 | 0 | 0 | 0 | 20 |
| Brewer’s yeast slurry | 310 | 147 | 0 | 0 | 0 | 0 |
| Soybean meal | 35 | 35 | 35 | 175 | 0 | 0 |
| Cottonseed meal | 255 | 298 | 298 | 0 | 0 | 350 |
| Cassava chips flour | 200 | 200 | 200 | 200 | 200 | 200 |
| Dried brewer’s yeast | 0 | 0 | 147 | 305 | 480 | 126 |
| Mineral and vitamin premix | 5 | 5 | 5 | 5 | 5 | 5 |
| Methionine | 2 | 2 | 2 | 2 | 2 | 2 |
| Red palm oil | 43 | 43 | 43 | 43 | 43 | 43 |
| Total | 1000 | 1000 | 1000 | 1000 | 1000 | 1000 |

[5]- Formulas have been developed based on the proximate composition, expressed on dry matter basis of ingredients.
Source: Our Data (2020).

release free gossypol during digestion (Noftsger et al., 2000), consequently limiting its use. Thus, Ashiru et al. (2015) recommended that the incorporation of cottonseed meal in the feeds of African catfish juveniles should not exceed the range of 200 to 300 g $kg^{-1}$.

Moringa leaf meal also demonstrated a promising nutritional profile for feed formulation. Moringa, a fast-growing plant widely available in the tropics and subtropics, has been recognized by several authors (Abo-State et al., 2014) for its considerable potential as an ingredient for animals and fish. Gbadamosi and Osungbemiro (2016) suggested that Moringa leaf meal can be included in catfish feed up to 100 g $kg^{-1}$ in replacement of fishmeal. Another advantage of Moringa is its antibacterial properties. Hammed et al. (2015) demonstrated that *C. gariepinus* infected with *Aeromonas* spp. bacteria can be effectively treated with Moringa leaf extract at a 50% concentration without adverse effects.

Cassava chips emerged as the most promising source of starch. Cassava offers several advantages compared to other carbohydrate sources, particularly other root crops. Firstly, over 85% of root dry matter consists of highly digestible starch. Secondly, cassava starch has excellent agglutination properties, making it particularly suitable for fish feed as it can replace other, more expensive artificial agglutinants. Carbohydrates (starches and sugars) are generally the cheapest sources of energy for fish diets. They are included in aquaculture diets to reduce feed costs and for their binding activity during extrusion to manufacture floating feeds. The cooking of starch during the extrusion process enhances its biological availability to fish (Craig et al., 2017).

Palm oil, although not ranked, was still used in the formulations because it is a common source of lipids in fish feeds (Adéyèmi et al., 2020). Besides its ample availability, lower cost, and sustainable production, its low polyunsaturated fatty acid content and relatively high amount of vitamin E make it the vegetable oil of choice for formulating fish feeds in tropical countries (Ng et al., 2003; Babalola and Apata, 2012). According to these authors, red palm oil can effectively replace fish oil in the diet of fish without compromising fish growth and feed efficiency.

## Feed formulation and characteristics

The major criteria in fish feed formulation are nutrient requirements and the costs of ingredients. Achieving the protein target of 420 g $kg^{-1}$ for adequate growth presents a significant challenge when formulating feed for catfish. Six feed formulas (Feed 1-Feed 6) were developed using selected ingredients, as shown in Table 4. One approach to formulating cost-effective yet highly nutritious fish feeds is by combining various protein by-products (Rawles et al., 2011).

Therefore, the objective of our study was to determine the best combination of diverse protein sources (such as fishmeal, trash fish meal, garden snail meal, poultry viscera meal, brewer’s yeast, soybean meal, and cottonseed meal) to formulate multi-protein, affordable, and nutritious fish feeds. Brewer’s yeast was used in its wet form (slurry) in two formulas (Feed 1 and Feed 2) and in its dried form in four formulas (Feed 3, Feed 4, Feed 5, and Feed 6).

Feed 2 contained less brewers’ yeast (147 g $kg^{-1}$) than Feed 1 (310 g $kg^{-1}$), with the difference compensated by fishmeal (120 g $kg^{-1}$). Feed 6 included garden snail meal (10 g $kg^{-1}$) and poultry viscera meal (20 g $kg^{-1}$), which were absent in the other feeds. In Feed 3, Feed 4, and Feed 5, the inclusion of dried brewer’s yeast was increased from 147 g $kg^{-1}$ (Feed 3) to 480 g $kg^{-1}$ (Feed 5).

**Table 5.** Nutritional composition (g kg$^{-1}$ DM basis) and cost (US $ kg$^{-1}$) of formulated feeds.

| Parameter | Feed 1 | Feed 2 | Feed 3 | Feed 4 | Feed 5 | Feed 6 | Control feed |
|---|---|---|---|---|---|---|---|
| **Software predicted values** | | | | | | | |
| Moisture | 342.0 | 217.7 | 96.5 | 78.3 | 68.3 | 97.0 | |
| Protein | 254.2 | 311.8 | 364.5 | 396.8 | 391.4 | 361.5 | |
| Fat | 62.2 | 71.6 | 73.4 | 75.9 | 73.3 | 71.9 | *No predicted value* |
| Carbohydrates | 278.6 | 289.9 | 353.7 | 366.8 | 390.0 | 359.0 | |
| Cost | 0.46 | 0.53 | 0.75 | 1.08 | 1.25 | 0.74 | |
| **Values from chemical analysis** | | | | | | | |
| Moisture | $115.0 \pm 11.9^{d}$ | $99.5 \pm 15.6^{f}$ | $155.6 \pm 3.6^{bc}$ | $172.7 \pm 0.0^{ab}$ | $178.2 \pm 10.1^{a}$ | $148.2 \pm 11.4^{c}$ | $73.7 \pm 1.2^{e}$ |
| Crude protein * | $389.6 \pm 16.5^{ab}$ | $370.4 \pm 17.3^{c}$ | $366.6 \pm 17.6^{c}$ | $404.7 \pm 0.9^{ab}$ | $366.3 \pm 13.8^{c}$ | $377.3 \pm 14.1^{ab}$ | $424.4 \pm 2.1^{a}$ |
| Crude fat ** | $84.2 \pm 0.41^{b}$ | $111.3 \pm 0.8^{a}$ | $106.5 \pm 5.5^{a}$ | $99.4 \pm 1.5^{ab}$ | $103.7 \pm 1.4^{a}$ | $103.5 \pm 9.1^{a}$ | $84.3 \pm 1.1^{b}$ |
| Ash | $80.7 \pm 5.8^{c}$ | $112.3 \pm 4.7^{a}$ | $96.9 \pm 1.7^{b}$ | $84.6 \pm 2.0^{c}$ | $81.7 \pm 2.0^{c}$ | $97.2 \pm 3.4^{b}$ | $71.3 \pm 0.6^{d}$ |
| Carbohydrates * | $330.5 \pm 30.1^{a}$ | $306.5 \pm 36.8^{ab}$ | $274.3 \pm 17.4^{ab}$ | $238.6 \pm 2.6^{b}$ | $270.0 \pm 23.3^{ab}$ | $273.7 \pm 3.0^{ab}$ | $346.2 \pm 0.8^{a}$ |
| Gross energy (kJ/g) | $19.30 \pm 0.23^{b}$ | $19.47 \pm 0.19^{b}$ | $18.64 \pm 0.04^{c}$ | $18.77 \pm 0.01^{c}$ | $18.45 \pm 0.03^{de}$ | $18.80 \pm 0.06^{c}$ | $20.50 \pm 0.03^{a}$ |

The values of chemical analysis are the averages of 4 repetitions. Mean values within a row having different superscripts are significantly different at $p < 0.05$.1 US $ = 580 Fcf.
Source: Our Data (2020).

Additionally, Feed 4 and Feed 5 did not contain cottonseed meal, and Feed 5 omitted soybean meal. Trash fish and dried brewer's yeast were the only protein sources in Feed 5.

The nutrient contents of the formulated feeds, as predicted by the software and analyzed chemically after production, along with their costs, are presented in Table 5. Feed 5 was the most expensive formulation (1.25 US$ kg$^{-1}$), while Feed 1 was the cheapest (0.46 US$ kg$^{-1}$). This cost estimation is based solely on ingredient prices and excludes other production expenses. Analysis of Variance on measured values indicated a significant difference between all six formulated and control feeds in terms of crude protein (p = 0.023), crude fat (p = 0.002), ash (p = 0.000), carbohydrate (p = 0.014) contents, and gross energy (p = 0.000).

Protein content in fish feed is crucia l in aquaculture, as adequate dietary protein significantly influences fish growth, survival, and feed cost. The protein content of formulated feeds ranged from 366 g kg$^{-1}$ (Feed 5) to 405 g kg$^{-1}$ (Feed 4).

The protein contents of Feed 1 (390 g kg$^{-1}$), Feed 4 (405 g kg$^{-1}$), and Feed 6 (377 g kg$^{-1}$) did not significantly differ from the control feed (424 g kg$^{-1}$). These top three feeds' protein values are also within the dietary protein range of 380 to 420 g kg$^{-1}$ recommended for adequate catfish growth (Jimoh et al., 2013). It's noteworthy that several authors have achieved good growth performance for African catfish using feed with lower protein contents ranging from 250 g kg$^{-1}$ to 380 g kg$^{-1}$, confirming that the efficiency of dietary protein for fish growth depends on both the quantity and quality of feed proteins (Gasco et al., 2016). In this respect, adequate catfish growth has been reported using feed containing (i) 388 g kg$^{-1}$ of protein from 200 g kg$^{-1}$ of mealworm (Ng et al., 2001), (ii) 356 g kg$^{-1}$ of crude protein by replacing 75% of fishmeal with cricket meal (Taufek et al., 2016), (iii) 300 g kg$^{-1}$ of crude protein by incorporating fermented silage from fish by-products (Soltan et al., 2008), and (iv) as little as 290 g kg$^{-1}$ protein by including 131 g kg$^{-1}$ of variegated grasshopper (Alegbeleye et al., 2012).

The crude fat contents of the feeds ranged between 84 g kg$^{-1}$ (Feed 1) and 111 g kg$^{-1}$ (Feed 2). All fat values in the formulated feeds roughly met the fat requirement (87 g kg$^{-1}$ to 170 g kg$^{-1}$) of catfish as reported by Ali and Jauncey (2005). The imported control feed had a fat content of 84 g kg-1, which was below the fat content of most of our feeds.

The ash contents of the feeds ranged from 81 g kg$^{-1}$ (Feed 1) to 112 g kg$^{-1}$ (Feed 2), all exceeding

**Table 6.** Essential amino acids (EAAs) and essential fatty acids (EFAs)[6] of formulated feeds in comparison to nutrient requirements (EAAs[7] and EFAs[8]) and of *Clarias gariepinus*.

| Nutrient | | Requirements (minimum) | Feed 1 | Feed 2 | Feed 3 | Feed 4 | Feed 5 | Feed 6 |
|---|---|---|---|---|---|---|---|---|
| EAAs (% of crude protein) | Arginine | 4.3 | 4.3 | 5.7 | 6.6 | 4.5 | 4.7 | 6.7 |
| | Histidine | 1.5 | 2.4 | 2.8 | 3.1 | 3.5 | 3.8 | 3.0 |
| | Isoleucine | 2.6 | 3.6 | 4.3 | 5.0 | 6.3 | 7.1 | 4.7 |
| | Leucine | 3.5 | 4.7 | 6.0 | 7.1 | 8.1 | 9.1 | 6.7 |
| | Lysine | 5.1 | 2.3 | 3.6 | 5.1 | 5.5 | 6.6 | 4.7 |
| | Methionine | 2.3 | 2.2 | 2.7 | 2.7 | 2.9 | 3.0 | 2.6 |
| | Threonine | 2.0 | 1.5 | 2.2 | 3.1 | 3.0 | 3.6 | 2.9 |
| | Tryptophan | 0.5 | 2.1 | 2.3 | 2.5 | 3.3 | 3.7 | 2.3 |
| | Valine | 3.0 | 3.2 | 4.2 | 5.3 | 6.0 | 6.8 | 5.0 |
| EFAs (g $kg^{-1}$ of feed) | 18:3n-3 | 10 | 1.3 | 1.4 | 1.4 | 1.6 | 1.1 | 1.4 |
| | 18:2n-6 | -- | 16.0 | 17.6 | 17.6 | 9.6 | 6.4 | 19.0 |
| Ratio Σn-3:Σn-6 | | 0.88 | 0.37 | 0.48 | 0.48 | 1.00 | 1.39 | 0.41 |

[6]- Values are estimated based on National Research Council (1993) and Robinson et al. (2001), [7]- Alegbeleye et al. (2012), [8]- Robinson and Li (1996).

the ash content of the control feed (71 g $kg^{-1}$). Carbohydrate contents ranged from 239 g $kg^{-1}$ (Feed 4) to 346 g $kg^{-1}$ (Control feed).

Controversy exists regarding the ability of catfish to properly utilize carbohydrates, although their natural feed may contain high levels of carbohydrates. The amount of carbohydrates in published Clarias feeds is substantial and ranged from 150 to 380 g $kg^{-1}$ (Ali and Jauncey, 2004). Consequently, the carbohydrate contents of the formulated feeds met the requirements of Clarias gariepinus. The highest gross energy value was 19.5 kJ $g^{-1}$ (Feed 2) and the lowest was 18.4 kJ $g^{-1}$ (Feed 5). All energy values in the formulated feeds were below that of the control feed (20.5 kJ $g^{-1}$) but higher than the required values (13 to17 kJ $g^{-1}$) reported by Van Weerd (1995) for catfish.

The calculated Essential Amino Acid (EAA) and Essential Fatty Acid (EFA) contents of the formulated feeds are given in Table 6. Overall, the EAA content of all formulated feeds satisfied the requirements for adequate growth of *C. gariepinus* except for lysine and threonine.

The lysine content of Feed 1 and Feed 2 and the threonine content of Feed 1 were lower than the required values as reported by Alegbeleye et al. (2012) for the catfish species. EFAs are essential for fish because they play important roles in cell synthesis, neural development, endocrine function and control, ionic regulation, immune function and reproduction (Glencross, 2009). The level of alpha-linolenic acid, an important EFA for fish, averaged 1.4 g $kg^{-1}$ in all formulated feeds and was lower than the required values of 10 to 20 g $kg^{-1}$ as reported by Robinson and Li (1996) for Clarias gariepinus. Linoleic acid (18:2n-6) was found in all formulated feeds at levels ranging from 6.4 to 19.0 g $kg^{-1}$ of feed.

## Clustering of extruded feeds based on their nutrient content

Principal Component Analysis (PCA) conducted on the nutrient data facilitated the grouping of the formulated feeds. The first two principal components account for 89.5% of the total variation, with PC1 explaining 68.7% and PC2 explaining 20.8% of the initial information. Figure 1 illustrates the clustering of formulated and control feeds based on their nutritional characteristics.

Protein, carbohydrates, and gross energy exhibit positive correlations with PC1, while fat and ash show negative correlations with PC1. The PCA resulted in the classification of feeds into three groups (G1, G2, and G3). Control Feed and Feed 1 in G1 display high protein, carbohydrate, and gross energy contents, alongside low ash and fat contents. Feeds in G2, encompassing Feed 2, Feed 3, and Feed 6, exhibit a negative correlation with PC1 and possess contrasting characteristics compared to the Control Feed and Feed 1. The high ash and low protein contents in these three feeds are attributed to the composition of the fishmeal available in Benin, which tends to be low in protein and high in ash content, as supported by Adéyèmi et al. (2020).

Feed 4 and Feed 5 in G3 demonstrate negative correlations with both PC1 and PC2. They are deficient in carbohydrates and do not offer more energy than Feed 1 and the Control Feed (G1). Moreover, they exhibit lower ash and fat contents compared to feeds in G2 (Feed 2 and Feed 3). Although Feed 4 boasts the highest protein content, Feed 1 was selected for an exploratory feeding trial due to its proximity to the control feed, that is, the commercially imported feed, in terms of their proximate compositions. Additionally, it had the lowest ingredient

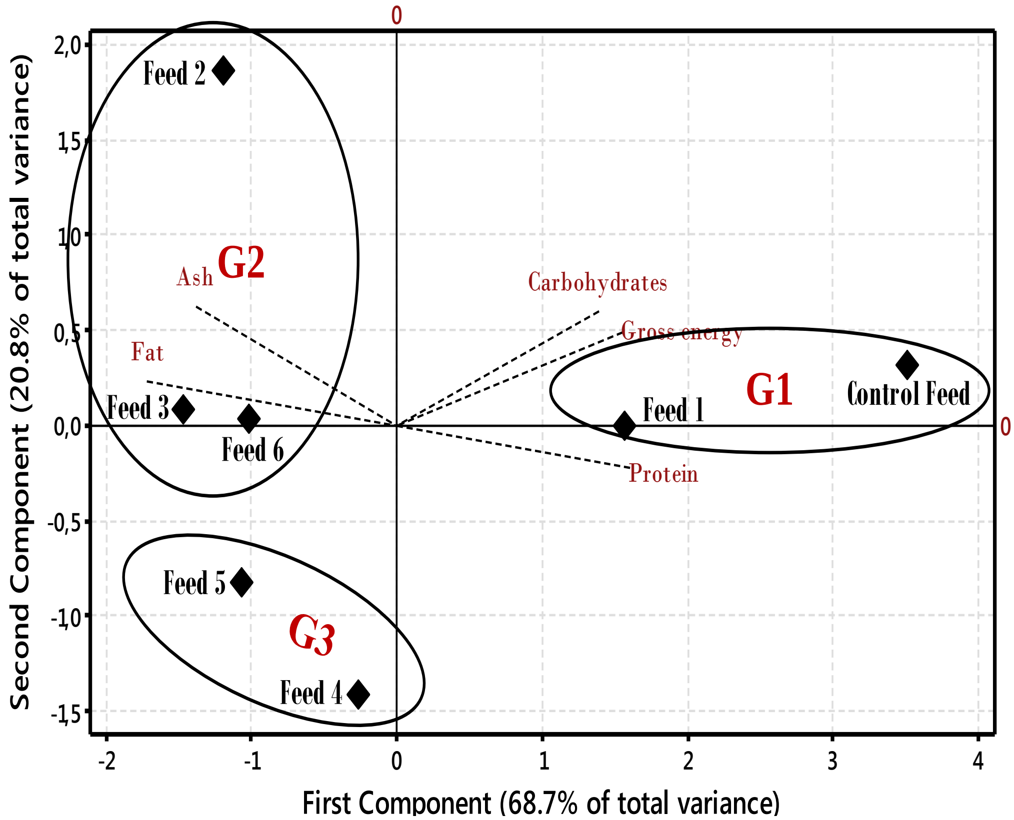


**Figure 1.** Biplot for clustering extrudates feed based on protein, fat, ash, carbohydrates and gross energy.

cost (0.46 US$ $kg^{-1}$).

## Water quality and growth performance of catfish in the exploratory feeding trial

The pH and temperature values of the water in the tanks were similar, ranging from 6.1 to 8.0 and 27.1 to 29.1, respectively. These data remained relatively stable and fell within the recommended range for the culture of *C. gariepinus* (pH: 6.5 to 9; temperature: 26 to 30°C) (Chabi et al., 2015). Dissolved Oxygen (DO) values were consistently low, varying from 0.30 mg/l to 5.8 mg/l, yet exhibited a consistent trend across all tanks. Furthermore, *C. gariepinus* has demonstrated resilience to marginal environmental conditions and can thrive in oxygen-deprived areas (Hecht, 1996).

No signs of disease due to suboptimal environmental conditions were observed during the trial. The growth performance and feed utilization of *C. gariepinus* fingerlings are presented in Table 7, with the weekly weight gain of fish depicted in Figure 2. Fish fed with the formulated feed displayed slower growth compared to those fed with the commercial control feed. By the end of the feeding trial, the highest individual fish weight (200.0 g) and the highest Specific Growth Rate (5.90% $day^{-1}$) were recorded for the fish fed with the commercial feed. Daily weight gain, feed intake, and protein intake for fish fed with the formulated feed (Feed 1) were 1.30 g, 88.6 g, and 34.5 g, respectively, while those for fish fed with the commercial feed were 3.44, 170.0 and 72.1 g, respectively, indicating notably higher values for the commercial feed.

This lower daily weight gain for fish fed Feed 1 was anticipated due to their reduced daily feed intake. However, the feed conversion ratio (FCR) and the protein efficiency ratio (PER) of both feeds were relatively similar. The FCR (1.25) and the PER (2.05) of Feed 1 were within an acceptable range when compared with previous studies, such as the FCR of 1.1 reported by Alofa et al. (2016) and the PER of 2.31 reported by Goda et al. (2007), suggesting that the slow growth may be attributed to reduced feed intake rather than the feed formulation itself.

Ashiru et al. (2015) suggested that high amounts (greater than 200 g $kg^{-1}$) of cottonseed meal in the feed may decrease feed intake and compromise fish growth.

Factors influencing feed intake and appetite warrant further investigation, and the use of attractants may potentially alleviate this limitation.

**Table 7.** Growth performance and feed utilization of *Clarias gariepinus* fed experimental feeds

| **Parameter** | | **Experimental feeds** | | ***p-value*** |
|---|---|---|---|---|
| | | **Feed 1** | **Commercial feed** | |
| Growth performance | Initial mean weight (g/fish) | 7.33 | 7.33 | -- |
| | Fish survival rate (%) *** | 91.67 ±0.0[b] | 99.44 ± 0.96[a] | 0000 |
| | Final mean weight (g/fish) *** | 80.0 ± 2.41[b] | 200.0 ± 1.80[a] | 0.000 |
| | Weight gain (g/fish/day) *** | 1.30 ± 0.04[b] | 3.44 ± 0.02[a] | 0.000 |
| | Specific growth rate (%/day) *** | 4.27 ± 0.05[b] | 5.90 ± 0.01[a] | 0.000 |
| Feed utilization | Feed intake (g/fish/day) *** | 88.57 ± 4.82[b] | 170.0 ± 6.07[a] | 0.000 |
| | Protein intake (g/fish/day) *** | 34.46 ± 1.78[b] | 72.14 ± 2.50[a] | 0.000 |
| | Feed conversion ratio*** | 1.25 ± 0.03[a] | 0.83 ± 0.01[b] | 0.000 |
| | Protein efficiency ratio*** | 2.05 ± 0.05[b] | 2.84 ± 0.06[a] | 0.000 |

Values are means of three replications.
Source: Our Data (2020).

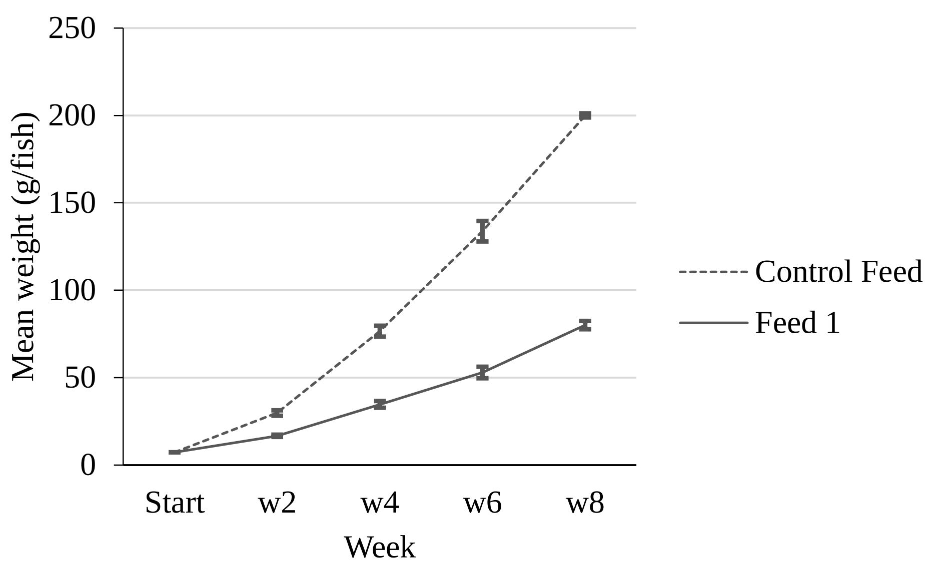


**Figure 2.** Weekly weight gain of *C. gariepinus* during the feeding trial.

## Conclusion

This study demonstrates that trash fish meal, soybean meal, and brewer's yeast are valuable protein sources for feed formulation for African catfish. High amounts of these ingredients can be utilized in catfish feed formulations, indicating the feasibility of formulating fish feed in Benin with sufficient nutritive value comparable to imported fish feed. However, the most promising new fish feed formula exhibited relatively low feed intake and consequent poor growth of catfish compared to the commercial feed control. To address this issue, reducing the high amount of cottonseed meal to less than 200 g $kg^{-1}$ in Feed 1 and incorporating attractants may stimulate fish appetite and increase feed intake. Further characterization of ingredients and formulated feeds, including digestibility, essential amino and fatty acid profiles, is recommended for fine-tuning feed formulation. Additionally, additional research is necessary on the physical properties of feeds, such as durability and floatability, to enhance feed intake and achieve feed performance comparable to that of commercially imported fish feed.

## CONFLICT OF INTERESTS

The authors have not declared any conflict of interests.

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