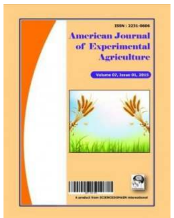


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# Growth Response and Nutrient Utilization of *Clarias gariepinus* Juveniles Fed Differently Processed Sunflower (*Helianthus annuus* Linnaeus) Seed Meal-Based Diets


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***Authors' contributions***

*This work was carried out by three authors. Authors AEF and SAA designed the study and wrote the protocol, author AEF supervised the study. Author SAA performed the feeding trial and the statistical analysis as well as managed the literature searches. Author SEO wrote the protocol, managed the literature searches and wrote the first draft of the manuscript. All authors read and approved the final manuscript.*


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## ABSTRACT


A 10-week feeding trial was conducted in 15 plastic tanks (60 × 45× 30 cm) to assess the performance of *Clarias gariepinus* juveniles fed diets containing sunflower seed meals processed by different techniques. Five diets were formulated at 40% crude protein content containing no sunflower meal (control) or 50% soybeans and 50% sunflower meal. The sunflower seed meals were processed using four methods: (a) Boiled Sunflower Seed Meal (BSSM); cooked, oven-dried and ground, (b) Roasted Sunflower Seed Meal (RSSM); roasted, cooled and ground, (c) Mechanically-Extracted Sunflower Seed Meal (MSSM); screw-pressed, oven-dried and ground and (d) Solvent-Extracted Sunflower Seed Meal (SSSM); de-oiled, air-dried, oven-dried and ground. Each treatment was done in triplicate contained 20 fish (mean weight 30.01±0.01 g and length



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12.31±0.30 cm). Fish were fed twice daily at 5% of their body weight. Fish fed the BSSM diet performed better ($p<0.05$) than those fed others processed sunflower meal diets and the control diet. *Clarias gariepinus* fed BSSM had significantly higher mean weight gain, specific growth rate, feed conversion ratio, gross feed conversion efficiency and nitrogen metabolism of 52.16±2.83 g, 0.62±0.09 g, 0.47±0.02, 212.77±0.02 and 2155.50±0.04 respectively of all the treatments. The processed sunflower seed meals recorded significantly higher ($p<0.05$) values of mineral composition (sodium, calcium, potassium, magnesium, iron and phosphorus) compared to the control. The result from this study indicated that BSSM diet can partially replace soybean meal in the diets for *C. gariepinus* without compromising growth or nutrient utilization.

*Keywords: Clarias gariepinus; sunflower; processing methods; fish growth; experimental diets.*

## 1. INTRODUCTION

African catfish, *Clarias gariepinus* is a common cultured fish species in Nigeria and sub- Sahara Africa [1], due to its ability to tolerate a wide range of environmental conditions, high stocking densities under culture conditions, relatively fast growth rate and good quality meat. Feed formulations account for more than 50% of the total production costs in modern intensive aquaculture [2]. Fishmeal has been the main protein source in balanced fish diets as it is rich in essential amino acids and minerals and highly digestible by fish. However, its use is limited by high cost and availability [1].

Any reduction in feed costs would have a direct positive effect on profitability of aquaculture. Oilseed cakes and legume seeds are considered suitable as alternative dietary protein sources for fish feed and are available in sub-Saharan African on a large-scale [3]. Therefore, this has necessitated the need to focus on using less expensive, less competitive and readily available plant protein sources such as sunflower seed meal to replace soybean meal without reducing the nutritional quality of the feed. The sunflower (*Helianthus annuus* Linnaeus.) is a member of the family *compositae*, a large and successful family of flowering plants occurring throughout the World [4]. Sunflower probably originated from the South-west United States of America, that is, Mexico area where its seeds were early used for food. Archeologically explorations at several sites in North America have discovered evidence of sunflower cultivation, which dated back to 3000 BC at sites in Arizona and New Mexico, apparently by Red Indians. It was later introduced to and cultivated in many other parts of the World. *Helianthus annuus* is a tall herbaceous annual plant grown mainly for its seeds, oil and other derivatives. The two cultivars of sunflower grown commercially are the oil-seed and non-oilseed types [4].

Sunflower seeds are sold as in-shell seeds or as dehulled/decorticated kernels. There is scarce information on uses of sunflower in the diets of *Clarias gariepinus*. This study was conducted to assess the performance of *Clarias gariepinus* juveniles fed diets containing differently processed sunflower seed meals.

## 2. MATERIALS AND METHODS

The experiment was carried out using fifteen plastic tanks (60 × 45× 30 cm) for 10 weeks in the Department of Aquaculture and Fisheries Management Laboratory of the University of Ibadan, Nigeria. The water level in each tank was maintained at a depth of 0.45 m throughout the experiment and replaced every three days to maintain relatively uniform physico-chemical parameters and prevent fouling from feed residues. Well water from the Department of Aquaculture and Fisheries Management, University of Ibadan was used for the experiment. Each tank was well aerated using air stones and aerator pumps (Cosmos aquarium air pump, double type 3500 50 Hz, 2.5 w-3 w) [5]. The dissolved oxygen content and pH of the water were measured using dissolved oxygen meter (Jenway 3015 pH meter, 0.0 accuracy; Genway, Staffordshire, UK) after standardizing the meter and water temperature by a bulb thermometer (Paragon Scientific Ltd, Birkenhead, Wirral, UK).

There were five dietary treatments each having three replicates, with 20 fish each with a mean initial body weight and standard length of 30.01±0.01 g and 12.31±0.30 cm, respectively. The fish were weighed, distributed into experimental tanks and allowed to acclimatize for 14 days before the experiment. The experiment lasted for 10 weeks during which the fish were fed at 5% body weight twice daily. The diet per day was divided into two; 2.5% given in the morning by 8.00 – 9.00 am and 2.5% in the evening by 5.00 pm. Weight changes were recorded weekly and feeding rates adjusted to the new body weights.

Raw undehulled sunflower seeds used in the study were collected from the sunflower farm-plot of the Teaching and Research Farm of the University of Ibadan. The seeds were immediately spread on wide polythene sheets to ensure uniform solar drying and moisture content for two weeks. During drying, remnants such as flower stalks, receptacles and fragments of stems and leaves were completely removed. The seeds were then packaged and stored in air-tight plastic containers prior to processing

Sunflower seeds were processed using six methods: (1) raw undehulled sunflower seed meal (RUSSM) was ground in a Thomas Wiley milling machine. The resultant ground mash was then oven-dried at 60℃ for 6 hours in a Gallenkamp oven prior to proximate analysis; (2) raw dehulled sunflower seed meal (RSSM) was obtained manually after removing the kernels from the seed coat. The kernels obtained were ground in a Thomas Wiley miller and then oven-dried in a Gallenkamp oven at 60℃ for 6 hours prior to proximate analysis; (3) boiled sunflower seed meal (BSSM) was prepared at 100℃ for 15 minutes in a pressure cooker (Qlink model no. 9000), oven-dried in a Gallenkamp oven at 60℃ for 6 hours prior to proximate analysis; (4) roasted dehulled sunflower seed meal (RSSM) was prepared in a Gallenkamp electric cooker and roasted at 80℃ for 45minutes, cooled and ground prior to proximate analysis; (5) mechanically-extracted sunflower seed meal (MSSM) was dried in a Leec drying cabinet at 80℃ for 30 minutes as described by [6], the resultant ground paste was loaded into the receptacle of an improvised mechanical screw press and screw-pressed for 24 hours. The resultant cake was hand-crumbled and oven-dried at 60℃ for 6 hours in a Gallenkamp oven prior to proximate analysis. (6) solvent-extracted sunflower seed meal (SSSM) was dried in a Leec drying cabinet at 80℃ for 30 minutes as described by [6]. The resultant ground paste was de-oiled in a soxhlet apparatus by soaking it inside three litres of 90% petroleum spirit (B.P.: 60-80℃) for 12 hours. The resultant powdery meal was initially air-dried and sun-dried for 1 day before it was finally oven-dried at 60℃ for 6 hours in a Gallenkamp oven. This was intended to ensure a thorough removal of any residual solvent prior to proximate analysis.

Each diet mixture was treated separately and extruded through a ¼ mm die mincer of Hobart A-200T pelleting machine (Hobart GmbH, Rben-Bosch, Offenbug, Germany) to form noodle-like strands, which were mechanically broken into suitable sizes for the *C. gariepinus* juveniles. The pellets were sun-dried, packed in labelled polythene bags and stored in a cool dry place to prevent fungal growth. Sunflower served as full or partial replacement for soybean meal (Table 1).

**Table 1. Gross ingredient compositions of experimental diets (g/100g dry matter)**

| Ingredients | Diet 1 (Control) | Diet 2 (BSSM ) | Diet 3 (RSSM) | Diet 4 (MSSM) | Diet 5 (SSSM) |
|---|---|---|---|---|---|
| Sunflower seed meal | - | 18.54 | 18.68 | 19.18 | 19.55 |
| Soybean meal | 37.08 | 18.54 | 18.68 | 19.18 | 19.55 |
| Fishmeal | 19.10 | 19.10 | 19.10 | 19.10 | 19.10 |
| Groundnut cake | 18.45 | 18.45 | 18.45 | 18.45 | 18.45 |
| Yellow maize | 18.62 | 18.62 | 18.34 | 17.34 | 16.60 |
| Palm oil | 0.75 | 0.75 | 0.75 | 0.75 | 0.75 |
| Bone meal | 1.00 | 1.00 | 1.00 | 1.00 | 1.00 |
| Oyster shell | 0.50 | 0.50 | 0.50 | 0.50 | 0.50 |
| Vitamin/mineral premix | 2.00 | 2.00 | 2.00 | 2.00 | 2.00 |
| Salt | 0.50 | 0.50 | 0.50 | 0.50 | 0.50 |
| Cassava starch | 1.00 | 1.00 | 1.00 | 1.00 | 1.00 |
| Chromic oxide | 0.50 | 0.50 | 0.50 | 0.50 | 0.50 |
| **Total (g)** | **100.00** | **100.00** | **100.00** | **100.00** | **100.00** |

*BSSM – Boiled sunflower seed meal, RSSM– Roasted sunflower seed meal, MSSM– Mechanically – extracted sunflower seed meal, SSSM– Solvent-extracted sunflower seed meal*

## 2.1 Biological Evaluation

Mean weight gain = [final mean weight (g) – initial mean weight (g)]

Mean weekly weight gain (MWWG)

$$MWWG\ (g/week) = W_f – W_i / n$$

Where:

$W_f$ = final fish weight at the end of the experiment.
$W_i$ = initial fish weight at the beginning of the experiment.
n = number of weeks (experimental duration).

Average daily growth (ADG)

ADG (mg/day) = Mean weight gain (in mg)/ Duration of feeding trials (days)

Relative growth rate (RGR)

$$RGR\ (\%) = (W_f - W_i) \times 100 / W_i$$

Where,

$W_f$ = final fish weight gain at the end of the experiment.
$W_i$ = initial fish weight at the beginning of the experiment [7].

Specific growth rate (SGR) = (Log $W_f$ – Log $W_i$) x 100 / t (days)

Where,

Log $W_f$ = logarithm of the fish final weight gain.
Log $W_i$ = logarithm of the fish initial weight, t = experimental period in days

$$\text{Condition factor (k)} = 100W / L^3$$

Where:

W = weight of fish, L = length of fish

Feed conversion ratio (FCR) = Feed intake (g)/ Weight gain (g)

Gross feed conversion efficiency (GFCE) = 1/ FCR x 100

Protein efficiency ratio (PER) = Wet body weight gain (g)/ Crude protein fed

Protein productive value (PPV) = (Final fish body protein - initial body protein)/ Crude protein intake × 100

Survival rate (%) =initial number of fish stocked – mortality / initial number of fish stocked × 100

Protein intake = feed intake × percentage protein in diet /100

Apparent net protein utilization (ANPU) = $P_2$ – $P_1$ / PI x 100

Where:

$P_1$ = initial protein in fish carcass at the beginning of the experiment
$P_2$ = final protein in fish carcass at the end of the experiment
PI = protein intake (g of protein in 100g of diet/fish) [8]

Nitrogen metabolism (NM) =0.549 x ($W_i$ + $W_f$) t/ 2

Where,

$W_i$ = Initial mean weight of fish, $W_f$ = Final mean weight of fish,
t = Experimental period in days, 0.549 = Metabolism factor [9]

Apparent digestibility of crude protein (ADp)

ADp (%) = (1 – dietary chromic oxide)/ faecal chromic oxide x faecal protein/ dietary protein x 100

Apparent digestibility of energy (ADe)

ADe (%) = (1 – dietary chromic oxide)/ faecal chromic oxide x faecal energy dietary energy x 100

## 2.2 Analytical Methods

Sunflower seed meal, experimental diets and fish carcasses were analyzed for proximate composition before and after the experiment using the methods of the [10].

## 2.3 Statistical Analysis

One-way analysis of variance (ANOVA) was used to analyze the data obtained during the trial using Statistical Package for Social Sciences (SPSS version 15). Duncan multiple range test was used to compare differences among individual means. Correlation and regression analyses were used to investigate relationships between weight and length.

## 3. RESULTS

### 3.1 Proximate and Mineral Compositions and Anti-nutritional Factors of Differently Processed Sunflower Seed Meals

Proximate composition (crude protein, crude fibre, crude fat, ash content and nitrogen free extract) of sunflower are included in Table 2. The result shown that SSSM had the highest crude protein, and the lowest was found in RUSSM; while the anti-nutrients factors (tannins, oxalate and phytate) showed higher values in RUSSM and lower in processed sunflower seed meal.

### 3.2 Proximate Compositions and Gross Energy Contents (kcal/kg) of Processed Sunflower Seed Meal used in Diets Fed to *Clarias gariepinus* Juveniles for 70 Days

Proximate composition (crude protein, crude fibre, crude fat, ash content and nitrogen free extract) was analyzed and highest crude protein was recorded in RSSM and the lowest in control diet, the processed sunflower seed meal was significantly difference (p<0.05) from control diet while highest crude fibre was recorded in BSSM and the lowest in control diet (Table 3).

**Table 2. Proximate and mineral compositions and anti-nutritional factors of differently processed sunflower seed meals**

| Components | RUSSM (% ± std) | RSM (% ± std) | SSSM (% ± std) | RSSM (% ± std) | MSSM (% ± std) | BSSM (% ± std) |
|---|---|---|---|---|---|---|
| Dry matter | $91.54{\pm}0.47^{b}$ | $91.56{\pm}0.15^{b}$ | $93.55{\pm}0.08^{a}$ | $89.71{\pm}0.51^{d}$ | $90.56{\pm}0.05^{c}$ | $91.22{\pm}0.09^{b}$ |
| Crude protein | $27.02{\pm}0.20^{e}$ | $33.82{\pm}0.23^{cd}$ | $45.31{\pm}0.06^{a}$ | $37.02{\pm}0.17^{bc}$ | $40.80{\pm}1.00^{b}$ | $32.21{\pm}0.13^{d}$ |
| Crude fibre | $26.35{\pm}0.17^{a}$ | $17.93{\pm}0.03^{d}$ | $18.40{\pm}0.42^{d}$ | $23.94{\pm}0.48^{b}$ | $21.04{\pm}1.02^{c}$ | $14.19{\pm}0.19^{e}$ |
| Crude lipid | $12.40{\pm}0.53^{c}$ | $20.84{\pm}0.02^{b}$ | $6.45{\pm}0.06^{e}$ | $12.41{\pm}0.01^{c}$ | $10.50{\pm}0.03^{d}$ | $21.60{\pm}0.01^{a}$ |
| Nitrogen-free extract | $19.52{\pm}0.04^{a}$ | $13.06{\pm}1.00^{d}$ | $17.77{\pm}0.04^{b}$ | $11.32{\pm}0.05^{e}$ | $12.91{\pm}0.07^{d}$ | $14.27{\pm}0.27^{c}$ |
| Moisture | $8.46{\pm}0.23^{c}$ | $8.44{\pm}0.02^{c}$ | $6.45{\pm}0.23^{a}$ | $10.29{\pm}0.23^{a}$ | $9.44{\pm}0.02^{b}$ | $8.78{\pm}0.07^{c}$ |
| Ash | $5.46{\pm}0.03^{a}$ | $5.20{\pm}0.04^{a}$ | $4.96{\pm}0.38^{abc}$ | $4.54{\pm}0.09^{c}$ | $4.61{\pm}0.03^{bc}$ | $5.10{\pm}0.03^{ab}$ |
| Calcium | $0.33{\pm}0.01^{b}$ | $0.27{\pm}0.02^{c}$ | $0.22{\pm}0.01^{d}$ | $0.22{\pm}0.01^{d}$ | $0.17{\pm}0.02^{e}$ | $0.38{\pm}0.02^{a}$ |
| Iron | $0.43{\pm}0.02^{b}$ | $0.52{\pm}0.02^{a}$ | $0.26{\pm}0.02^{de}$ | $0.25{\pm}0.03^{e}$ | $0.29{\pm}0.02^{d}$ | $0.35{\pm}0.01^{c}$ |
| Sodium | $0.23{\pm}0.01^{a}$ | $0.17{\pm}0.02^{a}$ | $0.15{\pm}0.01^{a}$ | $0.10{\pm}0.01^{a}$ | $0.25{\pm}0.21^{a}$ | $0.19{\pm}0.02^{a}$ |
| Potassium | $2.12{\pm}0.03^{a}$ | $2.03{\pm}0.02^{b}$ | $1.95{\pm}0.02^{c}$ | $1.87{\pm}0.02^{d}$ | $2.01{\pm}0.01^{b}$ | $1.96{\pm}0.02^{c}$ |
| Phosphorus | $1.68{\pm}0.02^{b}$ | $1.72{\pm}0.02^{a}$ | $1.63{\pm}0.02^{c}$ | $1.66{\pm}0.02^{bc}$ | $1.57{\pm}0.02^{d}$ | $1.69{\pm}0.03^{ab}$ |
| Magnesium | $0.37{\pm}0.02^{a}$ | $0.22{\pm}0.02^{b}$ | $0.13{\pm}0.03^{d}$ | $0.15{\pm}0.03^{cd}$ | $0.17{\pm}0.01^{c}$ | $0.24{\pm}0.02^{b}$ |
| Manganese | $0.15{\pm}0.02^{a}$ | $0.13{\pm}0.02^{a}$ | $0.13{\pm}0.01^{a}$ | $0.13{\pm}0.03^{a}$ | $0.15{\pm}0.02^{a}$ | $0.14{\pm}0.01^{a}$ |
| Copper | $0.02{\pm}0.01^{ab}$ | $0.04{\pm}0.01^{a}$ | $0.01{\pm}0.01^{b}$ | $0.03{\pm}0.01^{ab}$ | $0.01{\pm}0.01^{b}$ | $0.01{\pm}0.01^{b}$ |
| Zinc | $0.12{\pm}0.02^{abc}$ | $0.09{\pm}0.02^{c}$ | $0.14{\pm}0.02^{a}$ | $0.13{\pm}0.02^{ab}$ | $0.11{\pm}0.01^{bc}$ | $0.12{\pm}0.01^{ab}$ |
| Tannins | $0.45{\pm}0.03^{a}$ | $0.39{\pm}0.04^{a}$ | $0.40{\pm}0.05^{a}$ | $0.21{\pm}0.03^{b}$ | $0.40{\pm}0.05^{a}$ | $0.28{\pm}0.05^{b}$ |
| Oxalate | $0.18{\pm}0.07^{a}$ | $0.17{\pm}0.05^{a}$ | $0.16{\pm}0.02^{a}$ | $0.13{\pm}0.02^{a}$ | $0.16{\pm}0.03^{a}$ | $0.11{\pm}0.01^{a}$ |
| Phytate | $0.16{\pm}0.04^{a}$ | $0.14{\pm}0.03^{a}$ | $0.11{\pm}0.03^{a}$ | $0.14{\pm}0.04^{a}$ | $0.13{\pm}0.02^{a}$ | $0.12{\pm}0.03^{a}$ |

*The letters a, b,c,d,e,etc indicate that mean values in each row with similar superscripts are not significantly different (p>0.05). RUSSM – Raw un-dehulled sunflower seed meal; RSM – Raw dehulled sunflower seed meal; SSSM– Solvent-extracted sunflower seed meal; RSSM -Roasted sunflower seed meal; MSSM Mechanically – extracted sunflower seed meal; BSSM – Boiled sunflower seed meal; std – standard deviation.*

**Table 3. Proximate compositions and gross energy contents (kcal/kg) of processed sunflower seed meal used in diets fed to *Clarias gariepinus* juveniles for 70 days**

| Parameters | Diet 1 (Control) | Diet 2 (BSSM ) | Diet 3 (RSSM) | Diet 4 (MSSM) | Diet 5 (SSSM) |
|---|---|---|---|---|---|
| Crude protein (%) | $39.17{\pm}0.21^{c}$ | $39.84{\pm}0.28^{b}$ | $40.86{\pm}0.39^{a}$ | $40.59{\pm}0.47^{a}$ | $40.27{\pm}0.06^{ab}$ |
| Crude lipid (%) | $3.71{\pm}0.04^{b}$ | $3.73{\pm}0.14^{b}$ | $3.91{\pm}0.14^{b}$ | $5.22{\pm}0.33^{a}$ | $4.37{\pm}0.74^{b}$ |
| Crude fibre (%) | $2.92{\pm}0.03^{a}$ | $3.06{\pm}0.05^{a}$ | $2.87{\pm}0.21^{a}$ | $2.94{\pm}0.21^{a}$ | $2.94{\pm}0.11^{a}$ |
| Ash (%) | $11.53{\pm}0.04^{b}$ | $12.48{\pm}0.20^{a}$ | $12.73{\pm}0.67^{a}$ | $12.17{\pm}0.58^{ab}$ | $12.62{\pm}0.53^{a}$ |
| Moisture (%) | $8.56{\pm}0.11^{c}$ | $9.14{\pm}0.16^{ab}$ | $9.09{\pm}0.22^{b}$ | $9.41{\pm}0.05^{a}$ | $8.89{\pm}0.20^{b}$ |
| Nitrogen -free extract (%) | $34.12{\pm}0.21^{a}$ | $31.77{\pm}0.14^{b}$ | $30.55{\pm}0.13^{bc}$ | $29.69{\pm}1.54^{c}$ | $30.93{\pm}0.03^{bc}$ |
| Gross energy (Kcal/kg) | $4627.00{\pm}3.00^{a}$ | $4598.67{\pm}3.51^{a}$ | $4517.00{\pm}2.00^{b}$ | $4516.67{\pm}3.50^{b}$ | $4601.67{\pm}4.50^{a}$ |
| Calorie/protein ratio | $118.14{\pm}0.92^{a}$ | $115.43{\pm}1.30^{b}$ | $110.56{\pm}1.57^{c}$ | $111.30{\pm}1.97^{c}$ | $114.27{\pm}0.22^{b}$ |

*The letters a,b,c,d,e,etc indicate that mean values in each row with similar superscripts are not significantly different (p>0.05)*

### 3.3 Carcass Compositions of *Clarias gariepinus* Juveniles Fed Diets Including Processed Sunflower Seed Meal for 70 Days

Carcass composition (crude protein, crude fibre, crude fat, ash content and nitrogen free extract) of the fish before and the experiment are presented in Table 4. Highest crude protein was recorded in control diet and the lowest in SSSM after the experiment. There was no significant difference among the treated sunflower seed meal but, significant difference was observed between the treated sunflower seed meal and the control diet. Also, highest crude fibre was recorded in MSSM and the lowest in SSSM.

### 3.4 Growth Performance and Nutrient Utilization Indices of *Clarias gariepinus* Juveniles Fed Diets Including Processed Sunflower Seed Meal Diets for 70 Days

As observed in Table 5, mean weight gain was higher in fish fed diet boiled sunflower seed meal (52.16 g) and lower in those fed diet solvent-extracted sunflower seed meal (35.91 g), Specific growth rate was higher in the fish fed diet boiled sunflower seed meal (0.62%) and lower in those fed diet solvent-extracted sunflower seed meal (0.49%). FCR was higher in the fish fed diet roasted and solvent-extracted sunflower seed meal diets (0.62) and lower in those fed diet boiled sunflower seed meal (0.47). These values were significantly different among the dietary treatments and. Mean weekly weight changes of *Clarias gariepinus* juveniles fed diets including processed sunflower seed meal are presented in Fig. 1.

### 3.5 Mineral Compositions of *Clarias gariepinus* Juveniles before and after the Experiment

Mineral composition (sodium, calcium, potassium, magnesium, iron and phosphorus) before and after the experiment are included in Table 6. The highest values of phosphorus were found in MSSM and the lowest in BSSM and RSSM, no significant difference were observed among the treatments. Some minerals had lower values after the experiment when compared to the values recorded before the experiment (Table 6).

### 3.6 Initial and Final Mean Values of Water Quality Parameters Obtained during the 70 Days Feeding Trials

The pH, dissolved oxygen (mg/L) and temperature (℃) throughout the experimental period are shown in Table 7. The three parameters ranged within very close limits. The final values significantly differed ($p<0.05$) from the initial values.

## 4. DISCUSSION

In the present study, processing of sunflower seed meals using different methods produced variations in the proximate analyses and nutrient compositions of the meals. These variations consequently led to different levels of digestibility and nutrient utilization by the experimental fish. These are similar to the findings of [11] who reported variation in proximate composition of different processed sunflower seed meal. This is also similar to the reports on the utilization of differently processed feed ingredients (soybeans, cassava peels, feather and chicken offal and cotton seed,) by [12-14,6] respectively. The different processing methods used in this study (boiling, roasting, mechanical and solvent extraction) reduced the levels of anti-nutritional factors (tannins, oxalate and phytate) present in the raw sunflower seeds: since the anti-nutritional factors were lower in all the processed sunflower seed meals than in raw sunflower seed meal. Similar reductions have been reported when African yam beans were treated using these methods [1].

The reduction in the level of tannins resulting from the various processing methods used in this study is in agreement with the recommended methods for the removal of condensed tannins which include dehulling the seeds to remove the tannin-rich outer layer, autoclaving or treatment with alkali [15]. [16] observed a reduction in the tannin content of sesame seed meal from 20 g to 10 g/kg after fermentation with lactic acid bacteria.

The values of oxalates obtained in the study support the report of [17] who recorded reduces the oxalic acid content of the seed after dehulling. Oxalates reduce the physiological value of calcium in the seed [18,19]. Also, the value of phytate obtained in this study was similar to the report of [20] who reported reduction in phytate of processed sunflower seed meal. The bio-availability of phosphorus for animals seems to depend on the level of phytate-splitting enzyme, phytase, in the intestinal tract.

Body protein levels increased in all fish, indicating that all diets supported fish growth. Nevertheless, differences show that the whole body proximate composition was influenced by the method of processing sunflower meals. This result was in support with the report of [1] who recorded increased in protein levels of *C. gariepinus* juveniles fed diets differently processed African yam beans.

There was general improvement in growth performance and nutrient utilization of *Clarias gariepinus* fed diets differently processed sunflower seed meal. There were significant variations (p<0.05) in the mean weight gain of fish fed the four diets differently processed

**Table 4. Carcass compositions of *Clarias gariepinus* juveniles fed diets including processed sunflower seed meal before and after the 70 days trial**

| Parameters | Before | Diet 1 (Control) | Diet 2 (BSSM) | Diet 3 (RSSM) | Diet 4 (MSSM) | Diet 5 (SSSM) |
|---|---|---|---|---|---|---|
| Crude protein (%) | 59.88±1.40$^{c}$ | 66.54±1.21$^{a}$ | 62.79±0.40$^{b}$ | 62.97±1.60$^{b}$ | 63.43±0.55$^{b}$ | 62.59±0.87$^{b}$ |
| Crude lipid (%) | 5.14±0.08$^{d}$ | 12.56±0.37$^{c}$ | 13.87±0.20$^{a}$ | 14.12±0.01$^{a}$ | 13.08±0.20$^{b}$ | 14.14±0.18$^{a}$ |
| Crude fibre (%) | 1.39±0.08$^{a}$ | 1.19±0.03$^{b}$ | 1.21±0.07$^{b}$ | 1.24±0.06$^{b}$ | 1.25±0.08$^{b}$ | 1.16±0.07$^{b}$ |
| Ash (%) | 14.62±0.06$^{a}$ | 12.42±0.03$^{c}$ | 13.73±0.97$^{ab}$ | 13.81±0.79$^{ab}$ | 13.12±0.53$^{bc}$ | 12.60±0.85$^{bc}$ |
| Moisture (%) | 5.30±0.54$^{ab}$ | 5.96±0.92$^{a}$ | 5.77±0.12$^{ab}$ | 4.87±0.58$^{b}$ | 6.16±0.22$^{a}$ | 5.27±0.11$^{ab}$ |
| Nitrogen-free extract (%) | 13.68±1.05$^{a}$ | 2.67±0.59$^{b}$ | 2.98±0.82$^{b}$ | 2.98±1.45 | 2.98±0.11$^{b}$ | 4.32±1.75$^{b}$ |

*The letters a,b,c,d,e, etc indicate that mean values in each row with similar superscripts are not significantly different (p>0.05)*

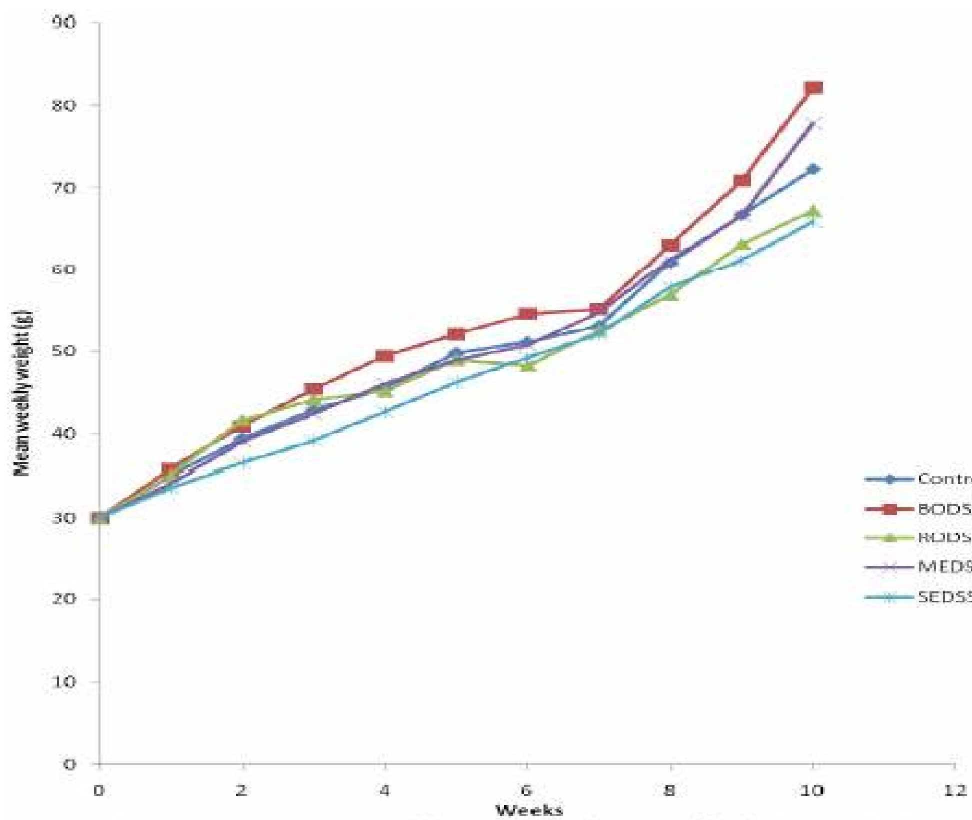


**Fig. 1. Mean weekly weight changes of *Clarias gariepinus* juveniles fed diets including processed sunflower seed meal diets**

sunflower seed meal. These occurred as a result of the improvement of the digestibility of the diets. This observation is similar to the findings of [21,8] who also recorded significant mean weight gain in *C. gariepinus* with higher digestibility values during their respective studies. Increase in weight gain in all experimental treatments indicated that the experimental fish were able to convert the protein in the feed to extra muscle in their bodies. The result of this study is in accord with the findings of [22] who reported higher growth of *Cirrhinus mrigala* fed diets including sunflower meal. Similarly, [23] reported that hybrid fingerlings (*Catla catla* x *Labeo rohita*) gained higher body weight and maximum total length when fed diets with sunflower seed meal. The highest (0.62%) and the lowest (0.49%) values of specific growth rate were recorded for boiled and mechanically-extracted sunflower seed meal diets respectively. These values are slightly lower than the 0.904% and 0.668% recorded for *Cyprinus carpio* fed fish meal-based and sunflower meal-based diets respectively [24]. Different processing methods (BSSM AND MSSM) were observed to produce significant differences in the values of specific growth rate when compare to the control diets. Feed conversion ratio (FCR) was the highest (0.62) in fish fed RSSM and SSSM diets and the lowest in those fed BSSM diet (0.47); this indicates a superior level of utilization of the BSSM diet by the fish. The lower FCR indicates better feed utilization by the fish and is justified by better growth performance of *C. gariepinus* fed BSSM diet. Apparent net protein utilization (ANPU) was 18.30% in fish fed the control diet. These values were significantly different in all the treatments. Fish fed MSSM diet had the highest ANPU value (7.20%) while those fed SSSM diet had the lowest (4.95%) among the processed sunflower meal diets. ANPU is a factor of the digestibility, utilization and quality of the protein fed to fish [25]. ANPU had been linked to superiority of feed and its protein quality [26] Therefore, fish fed MSSM diet had the highest productive protein value (3.03) apart from the control (7.71).

**Table 5. Growth performance and nutrient utilization indices of *Clarias gariepinus* juveniles fed diets including processed sunflower seed meal for 70 days**

| Parameters | Experimental Diets | | | | |
|---|---|---|---|---|---|
| | **Diet 1 (Control)** | **Diet 2 (BSSM)** | **Diets 3 (RSSM)** | **Diet 4 (MSSM)** | **Diet 5 (SSSM)** |
| Mean initial weight (g) | $30.01±0.01^{a}$ | $30.01±0.01^{a}$ | $30.01±0.00^{a}$ | $30.01±0.01^{a}$ | $30.01±0.01^{a}$ |
| Mean final weight (g) | $72.25±3.63^{ab}$ | $82.16±2.83^{a}$ | $67.26±1.16^{b}$ | $77.83±3.54^{ab}$ | $65.92±2.06^{b}$ |
| Mean weight gain (g) | $42.24±3.82^{ab}$ | $52.16±2.83^{a}$ | $37.25±1.25^{b}$ | $47.82±3.51^{ab}$ | $35.91±2.92^{b}$ |
| Percentage weight gain (%) | $140.75±12.73^{ab}$ | $173.81±9.42^{a}$ | $124.13±4.16^{b}$ | $159.35±18.34^{ab}$ | $119.66±9.75^{b}$ |
| Mean weekly weight gain (%) | $4.22±0.66^{ab}$ | $5.22±0.49^{a}$ | $3.73±0.22^{b}$ | $4.78±0.95^{ab}$ | $3.59±0.51^{b}$ |
| Mean initial length (cm) | $12.24±0.31^{a}$ | $12.40±0.21^{a}$ | $12.17±0.32^{a}$ | $12.10±0.10^{a}$ | $12.63±0.55^{a}$ |
| Mean final length (cm) | $22.47±0.35^{a}$ | $22.83±0.29^{a}$ | $21.63±0.61^{b}$ | $22.70±0.10^{a}$ | $21.63±0.47^{b}$ |
| Average daily growth (mg/day) | $603.43±54.63^{ab}$ | $745.14± 40.46^{a}$ | $532.14±17.82^{b}$ | $683.14±78.68^{ab}$ | $513.00±41.78^{b}$ |
| Relative growth rate (%) | $140.75±12.73^{ab}$ | $173.81±9.42^{a}$ | $124.13±4.16^{b}$ | $159.35±18.34^{ab}$ | $119.66±9.75^{b}$ |
| Specific growth rate (%) | $0.55±0.13^{ab}$ | $0.62±0.09^{a}$ | $0.50±0.05^{b}$ | $0.59±0.17^{ab}$ | $0.49±0.11^{b}$ |
| Feed conversion ratio | $0.56±0.02^{ab}$ | $0.47±0.02^{b}$ | $0.62±0.02^{a}$ | $0.50±0.02^{ab}$ | $0.62±0.02^{a}$ |
| Gross food conversion efficiency (%) | $178.57±0.02^{ab}$ | $212.77±0.02^{a}$ | $161.29±0.01^{b}$ | $200.00±0.02^{ab}$ | $161.29±0.01^{b}$ |
| Protein intake (g/100g diet/fish) | $0.95±0.04^{a}$ | $0.99±0.03^{a}$ | $0.92±0.02^{a}$ | $0.95±0.03^{a}$ | $0.90±0.02^{a}$ |
| Apparent net protein utilization (%) | $18.30±0.05^{a}$ | $5.60±0.02^{d}$ | $6.05±0.04^{c}$ | $7.20±0.04^{b}$ | $4.95±0.02^{e}$ |
| Productive protein value | $7.71±0.02^{a}$ | $2.26±0.02^{d}$ | $2.63±0.02^{c}$ | $3.03±0.03^{b}$ | $2.20±0.05^{e}$ |
| Nitrogen metabolism | $1964.90±0.02^{c}$ | $2155.50±0.04^{a}$ | $1869.04±0.02^{d}$ | $2072.15±0.08^{b}$ | $1843.29±0.01^{e}$ |
| Protein efficiency ratio | $1.06±0.03^{c}$ | $1.30±0.04^{a}$ | $0.93±0.02^{d}$ | $1.20 ± 0.04^{b}$ | $0.90±0.02^{d}$ |
| Condition factor (K) | $0.98±0.04^{a}$ | $1.01±0.02^{a}$ | $1.00±0.07^{a}$ | $0.94±0.08^{a}$ | $0.94±0.08^{a}$ |
| Survival rate (%) | $100.00±0.00^{a}$ | $96.67±1.67^{a}$ | $96.67±1.67^{a}$ | $98.33±1.67^{a}$ | $100.00±0.00^{a}$ |
| Apparent digestibility of crude protein (%) | $82.31±0.01^{c}$ | $81.50±0.03^{d}$ | $80.69±0.03^{e}$ | $85.91±0.01^{a}$ | $82.39±0.01^{b}$ |
| Apparent digestibility of gross energy (%) | $98.97±0.01^{a}$ | $98.98±0.01^{a}$ | $98.97±0.02^{a}$ | $98.97±0.01^{a}$ | $98.97±0.01^{a}$ |

*The letters a,b,c,d,e, etc indicate that mean values in each row with similar superscripts are not significantly different (p>0.05)*

**Table 6. Mineral compositions of *Clarias gariepinus* juveniles fed processed sunflower seed meal diets for 70 days**

| Parameters | Before | Diet 1 (Control ) | Diet 2 (BSSM ) | Diet 3 (RSSM) | Diet 4 (MSSM) | Diet 5 (SSSM) |
|---|---|---|---|---|---|---|
| Na (%) | 0.37±0.05$^{ab}$ | 0.35±0.01$^{b}$ | 0.41±0.03$^{a}$ | 0.30±0.01$^{c}$ | 0.23±0.02$^{d}$ | 0.26±0.02$^{d}$ |
| Ca (%) | 6.06±0.19$^{a}$ | 6.11±0.05$^{a}$ | 6.18±0.21$^{a}$ | 5.63±0.05$^{b}$ | 6.14±0.35$^{a}$ | 6.39±0.18$^{a}$ |
| K (%) | 0.29±0.02$^{a}$ | 0.20±0.02$^{c}$ | 0.24±0.03$^{abc}$ | 0.22±0.03$^{c}$ | 0.28±0.02$^{ab}$ | 0.23±0.06$^{bc}$ |
| Mg (%) | 0.31±0.02$^{ab}$ | 0.25±0.01$^{c}$ | 0.30±0.01$^{ab}$ | 0.28±0.04$^{bc}$ | 0.25±0.04$^{c}$ | 0.34±0.01$^{a}$ |
| Fe (mg/kg) | 404.00±8.00$^{ab}$ | 409.00±17.00$^{a}$ | 385.67±1.53$^{b}$ | 388.67±7.51$^{b}$ | 398.67±9.50$^{ab}$ | 409.67±7.51$^{a}$ |
| P (%) | 3.75±0.08$^{a}$ | 3.62±0.17$^{a}$ | 3.57±0.03$^{a}$ | 3.57±0.09$^{a}$ | 3.67±0.06$^{a}$ | 3.65±0.23$^{a}$ |

*The letters a,b,c,d,e,etc indicate that mean values in each row with similar superscripts are not significantly different (p>0.05)*

**Table 7. Initial and final mean values of water quality parameters obtained during the 70 days feeding trials**

| Diets | Parameter | Initial | Week 2 | Week 4 | Week 6 | Week 8 | Week 10 | Mean |
|---|---|---|---|---|---|---|---|---|
| Control | Temp (°C) | 27.05±0.00 | 27.52±0.02 | 27.53±0.01 | 27.67±0.02 | 28.01±0.00 | 28.55±0.01 | 27.72±0 .47 |
| | Ph | 6.81±0.00 | 7.47±0.05 | 7.46±0.01 | 7.52±0.02 | 7.51±0.0 2 | 7.59±0.01 | 7.39±0.26 |
| | DO (mg/l) | 5.39±0.03 | 5.42±0.01 | 5.40±0.00 | 5.39±0.01 | 5 .41±0.00 | 5.40±0.00 | 5.40±0.01 |
| BSSM | Temp (°C) | 27.07±0.02 | 27.52±0.03 | 27.52±0.01 | 27.65±0.02 | 28.12±0.00 | 28.05±0.05 | 27.66±0 .35 |
| | pH | 6.64±0.03 | 7.53±0.05 | 7.54±0.04 | 7.56±0.03 | 7.58±0.0 2 | 7.56±0.01 | 7.40±0.34 |
| | DO (mg/l) | 5.41±0.00 | 5.41±0.01 | 5.40±0.00 | 5.40±0.00 | 5 .39±0.01 | 5.40±0.00 | 5.40±0.01 |
| RSSM | Temp (°C) | 27.02±0.01 | 27.52±0.01 | 27.52±0.03 | 27.65±0.01 | 28.07±0.00 | 28.05±0.04 | 27.64±0 .36 |
| | pH | 6.83±0.02 | 7.60±0.00 | 7.61±0.01 | 7.66±0.01 | 7.64±0.0 3 | 7.67±0.02 | 7.50±0.30 |
| | DO (mg/l) | 5.42±0.01 | 5.40±0.01 | 5.38±0.02 | 5.38±0.02 | 5 .40±0.00 | 5.41±0.01 | 5.40±0.01 |
| MSSM | Temp (°C) | 27.06±0.03 | 27.51±0.04 | 27.50±0.01 | 27.66±0.02 | 28.06±0.03 | 28.06±0.00 | 27.64±0 .35 |
| | pH | 6.55±0.05 | 7.83±0.01 | 7.76±0.03 | 7.77±0.06 | 7.79±0.0 1 | 7.84±0.05 | 7.59±0.47 |
| | DO (mg/l) | 5.41±0.01 | 5.37±0.02 | 5.38±0.02 | 5.41±0.01 | 5 .40±0.00 | 5.39±0.02 | 5.39±0.01 |
| SSSM | Temp (°C) | 27.21±0.00 | 27.51±0.01 | 27.50±0.05 | 27.67±0.03 | 28.05±0.01 | 28.05±0.00 | 27.67±0 .31 |
| | Ph | 6.85±0.02 | 7.80±0.00 | 7.80±0.01 | 7.79±0.02 | 7.75±0.0 3 | 7.76±0.03 | 7.63±0.37 |
| | DO (mg/l) | 5.41±0.01 | 5.36±0.03 | 5.39±0.02 | 5.40±0.01 | 5 .40±0.01 | 5.39±0.02 | 5.39±0.02 |

Protein Efficiency Ratio (PER) is a measurement of protein effectiveness to provide the essential amino acids needed by the fish as well as an index that had been associated with fat deposition in fish muscles [27]. Protein Efficiency Ratio (PER) showed significant differences among the treatments. Apart from the fish fed the control diet which had a protein efficiency ratio (PER) of 1.06, the highest PER value (1.30) was obtained in the fish fed BSSM diet and the least value (0.90) in those fed SSSM diets. The best body weight gain, specific growth rate, feed conversion ratio, gross feed efficiency ratio and nitrogen metabolism were recorded in *Clarias gariepinus* fed BSSM diet.

The survival rate of the control and SSSM treatments was 100% but survival in the RSSM and BSSM treatments was only 96.67±1.67%. These values were closely related and were not significantly different (p>0.05) among the treatments. However, the robustness and general well-being of the fish fed the sunflower seed meal diets are expressed by the condition factor (K), which did not significantly differ from the control (Table 5). Also in terms of biological parameters, the solvent–extracted sunflower seed meal diet produced the lowest value, which could be due to the presence of thioglucosides. Anti-nutritional factors such as tannin, oxalate acid, saponin, trypsin inhibitor, thioglucosides and phytate in sunflower seed meals can reduce protein digestibility, thereby reducing the nutritive value in relation to the availability of amino acids and growth performance [28]

Apparent digestibility of crude protein was highest (85.91%) in the fish fed MSSM diet and lowest in RSSM diet (80.69%), the values were significantly different among all dietary treatments and fell within the recommended range as reported by [29]. Apparent digestibility of gross energy was generally high in all the treatments and showed no significant difference. Since protein synthesis and maximum energy intake contribute immensely to maximum growth, it could be inferred that the amount of energy ingested and digested was the required quantity by *Clarias gariepinus* for the best conversion of feed into flesh. Apparent digestibility is mainly used to describe how efficiently feeds or feed ingredients are being digested and how much of their nutrient composition can be made available to fish for maintenance and growth [30].

Minerals are important for normal growth and serve as co-factors in several metabolic reactions of animals including fish. A reduction of any of them from the diet will negatively affect growth. In the present study, the processed sunflower seed meal diets recorded significant ($p<0.05$) values of mineral deposition of sodium, calcium, potassium, magnesium, iron and phosphorus. The highest value of phosphorus, iron, magnesium and calcium were recorded in SSSM. Water quality parameters are given in Table 7 and were within recommended limits for warm-water fishes [31,32]. The differences among sexes were not considered in the study.

## 5. CONCLUSION

In conclusion, this study showed that boiling, roasting, mechanical and solvent extraction methods ultimately increased crude protein content and simultaneously reduced the levels of anti-nutrients (tannins, oxalate and phytate) in the processed sunflower seed meal. *Clarias gariepinus* fed differently processed sunflower seed meal diets has demonstrated that sunflower seed meal should be included in the list of conventional aqua-feed ingredients. It is therefore recommended that supplementation of BSSM in the diets for *Clarias gariepinus* could boost aquaculture production and should therefore be further investigated on inclusion levels.

## COMPETING INTERESTS

Authors have declared that no competing interests exist.

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