

GROWTH RESPONSE AND NUTRIENT UTILIZATION OF *Clarias gariepinus* JUVENILES FED DIFFERENTLY PROCESSED SUNFLOWER (*Helianthus annuus* Linnaeus) SEED MEAL-BASED DIETS.

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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±0.01g and length 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±2.83g, 0.62±0.09g, 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, Fish growth, Experimental diets, Mineral composition

30 1. INTRODUCTION

31 African catfish, *Clarias gariepinus* is a common cultured fish species in Nigeria and
32 sub- Sahara Africa (Olaifa and Bello, 2011), due to its ability to tolerate a wide range of
33 environmental conditions, high stocking densities under culture conditions, relatively fast
34 growth rate and good quality meat. Feed formulations account for more than 50% of the total
35 production costs in modern intensive aquaculture (Ibrahem *et al.*, 2010). Fishmeal has been
36 the main protein source in balanced fish diets as it is rich in essential amino acids and
37 minerals and highly digestible by fish. However, its use is limited by high cost and availability
38 (Olaifa and Bello, 2011).

39 Any reduction in feed costs would have a direct positive effect on profitability of
40 aquaculture. The sunflower (*Helianthus annuus* Linnaeus.) is a member of the family
41 *compositae*, a large and successful family of flowering plants occurring throughout the World
42 (Iqbal, 2001). Sunflower probably originated from the South-west United States of America,
43 that is, Mexico area where its seeds were early used for food. It was later introduced to and
44 cultivated in many other parts of the World. *Helianthus annuus* is a tall herbaceous annual
45 plant grown mainly for its seeds, oil and other derivatives. The two cultivars of sunflower
46 grown commercially are the oil-seed and non-oilseed types (Iqbal, 2001).

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

51 2. MATERIAL AND METHODS

52 The experiment was carried out using fifteen plastic tanks (60 × 45× 30 cm) for 10
53 weeks in the Department of Aquaculture and Fisheries Management Laboratory of the
54 University of Ibadan, Nigeria. The water level in each tank was maintained at a depth of 0.45
55 m throughout the experiment and replaced every three days to maintain relatively uniform
56 physico-chemical parameters and prevent fouling from feed residues. Well water from the

57 Department of Aquaculture and Fisheries Management, University of Ibadan was used for
58 the experiment. Each tank was well aerated using air stones and aerator pumps (Cosmos
59 aquarium air pump, double type 3500 50Hz, 2.5w-3w) (Lawson 1995). The dissolved oxygen
60 content and pH of the water were measured using dissolved oxygen meter (Jenway 3015 pH
61 meter, 0.0 accuracy; Genway, Staffordshire, UK) after standardizing the meter and water
62 temperature by a bulb thermometer (Paragon Scientific Ltd, Birkenhead, Wirral, UK).

63 There were five dietary treatments each having three replicates, with 20 fish each
64 with a mean initial body weight and standard length of 30.01 ± 0.01 g and 12.31 ± 0.30 cm,
65 respectively. The fish were weighed, distributed into experimental tanks and allowed to
66 acclimatize for 14 days before the experiment. The experiment lasted for 10 weeks during
67 which the fish were fed at 5% body weight (in two equal portions of 2.5%) twice daily. Weight
68 changes were recorded weekly and feeding rates adjusted to the new body weights.

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

75 Sunflower seeds were processed using six methods: (1) raw unde-hulled sunflower
76 seed meal (RUSSM) was ground in a Thomas Wiley milling machine. The resultant ground
77 mash was then oven-dried at 60°C for 6 hours in a Gallenkamp oven prior to proximate
78 analysis; (2) raw dehulled sunflower seed meal (RSSM) was obtained manually after
79 removing the kernels from the seed coat. The kernels obtained were ground in a Thomas
80 Wiley miller and then oven-dried in a Gallenkamp oven at 60°C for 6 hours prior to proximate
81 analysis; (3) boiled sunflower seed meal (BSSM) was prepared at 100°C for 15 minutes in a
82 pressure cooker (Qlink model no. 9000), oven-dried in a Gallenkamp oven at 60°C for 6
83 hours prior to proximate analysis; (4) roasted dehulled sunflower seed meal (RSSM) was

84 prepared in a Gallenkamp electric cooker and roasted at 80⁰C for 45minutes, cooled and
85 ground prior to proximate analysis; (5) mechanically-extracted sunflower seed meal (MSSM)
86 was dried in a Leec drying cabinet at 80⁰C for 30 minutes as described by Alegbeleye
87 (2005), the resultant ground paste was loaded into the receptacle of an improvised
88 mechanical screw press and screw-pressed for 24 hours. The resultant cake was hand-
89 crumbled and oven-dried at 60⁰C for 6 hours in a Gallenkamp oven prior to proximate
90 analysis. (6) solvent-extracted sunflower seed meal (SSSM) was dried in a Leec drying
91 cabinet at 80⁰C for 30 minutes as described by Alegbeleye (2005). The resultant ground
92 paste was de-oiled in a soxhlet apparatus by soaking it inside three litres of 90% petroleum
93 spirit (B.P.: 60-80⁰C) for 12 hours. The resultant powdery meal was initially air-dried and
94 sun-dried for 1 day before it was finally oven-dried at 60⁰C for 6 hours in a Gallenkamp oven.
95 This was intended to ensure a thorough removal of any residual solvent prior to proximate
96 analysis.

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

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118 **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

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 120 BSSM – Boiled sunflower seed meal, RSSM– Roasted sunflower seed meal, MSSM–
 121 Mechanically – extracted sunflower seed meal, SSSM– Solvent-extracted sunflower seed
 122 meal
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124 **2.1 BIOLOGICAL EVALUATION**

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

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 127 Mean weekly weight gain (MWWG)

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$$\text{MWWG (g/week)} = \frac{W_f - W_i}{n}$$

129
 130 Where: W_f = final fish weight at the end of the experiment.

131 W_i = initial fish weight at the beginning of the experiment.

132 n = number of weeks (experimental duration).
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134 Average daily growth (ADG)

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$$\text{ADG (mg/day)} = \frac{\text{Mean weight gain (in mg)}}{\text{Duration of feeding trials (days)}}$$

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 137 Relative growth rate (RGR)

138
$$\text{RGR (\%)} = \frac{(W_f - W_i) \times 100}{W_i}$$

139 Where,

140 W_f = final fish weight gain at the end of the experiment.

141 W_i = initial fish weight at the beginning of the experiment

142 (Fasakin *et al.*, 2000)
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$$\text{Specific growth rate (SGR)} = \frac{(\text{Log } W_f - \text{Log } W_i) \times 100}{t \text{ (days)}}$$

145 Where, $\text{Log } W_f$ = logarithm of the fish final weight gain.

146 $\text{Log } W_i$ = logarithm of the fish initial weight, t = experimental period in days
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148
$$\text{Condition factor (k)} = \frac{100W}{L^3}$$

149 Where: W = weight of fish, L = length of fish
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151 Feed conversion ratio (FCR) = Feed intake (g) / Weight gain (g)
 152 Gross feed conversion efficiency (GFCE) = $1 / \text{FCR} \times 100$
 153 Protein efficiency ratio (PER) = Wet body weight gain (g)/ Crude protein fed
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 155 Protein productive value (PPV) =
 156 (Final fish body protein - initial body protein)/ Crude protein intake $\times 100$
 157
 158 Survival rate (%)
 159 =initial number of fish stocked – mortality / initial number of fish stocked $\times 100$
 160
 161 Protein intake = feed intake $\times$ percentage protein in diet /100
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 163 Apparent net protein utilization (ANPU) = $P_2 - P_1 / \text{PI} \times 100$
 164 Where: P_1 = initial protein in fish carcass at the beginning of the experiment
 165 P_2 = final protein in fish carcass at the end of the experiment
 166 PI = protein intake (g of protein in 100g of diet/fish) (Ali (2001)
 167
 168 Nitrogen metabolism (NM) = $0.549 \times (W_i + W_f) t / 2$
 169 Where, W_i = Initial mean weight of fish, W_f = Final mean weight of fish,
 170 t = Experimental period in days, 0.549 =Metabolism factor (Nwanna, 2003).
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172 2.2 ANALYTICAL METHODS

173 Sunflower seed meal, experimental diets and fish carcasses were analyzed for
 174 proximate composition before and after the experiment using the methods of the Association
 175 of Official Analytical Chemists (2005).

176 2.3 STATISTICAL ANALYSIS

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

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184 3. RESULTS

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186 3.1 Proximate and mineral compositions and anti-nutritional factors of 187 differently processed sunflower seed meals

188 Proximate composition (crude protein, crude fibre, crude fat, ash content and
189 nitrogen free extract) of sunflower seed meals was determined and are included in table 2.
190 The result shown that SSSM had the highest crude protein, and the and lowest was found in
191 RUSSM; while the anti-nutrients factors (tannins, oxalate and phytate) showed a higher
192 values in RUSSM and lower values were obtained in processed sunflower seed meal (table
193 2-).

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

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

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

198 RUSSM – Raw un-dehulled sunflower seed meal; RSM – Raw dehulled sunflower seed
199 meal; SSSM– Solvent-extracted sunflower seed meal; RSSM -Roasted sunflower seed
200 meal; MSSM Mechanically – extracted sunflower seed meal; BSSM – Boiled sunflower seed
201 meal; std – standard deviation.

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

Proximate composition (crude protein, crude fibre, crude fat, ash content and nitrogen free extract) ~~are shown in table 3. The 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. ~~The while~~ highest crude fibre was recorded in BSSM and the lowest in control diet (Table 3)

Table 3: Proximate compositions and gross energy contents (kcal/kg) of processed sunflower seed meal ~~used in~~ diets to 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) was analyzed of the fish before and after 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 ($p>0.05$) among the treated sunflower seed meal but, significant difference ($p<0.05$) 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

Table 4: Carcass compositions of *Clarias gariepinus* juveniles fed diets including processed sunflower seed meal for 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
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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).

3.4 Growth performance and nutrient utilization indices of *Clarias gariepinus*

juveniles fed diets including processed sunflower seed meal diets for 70 days

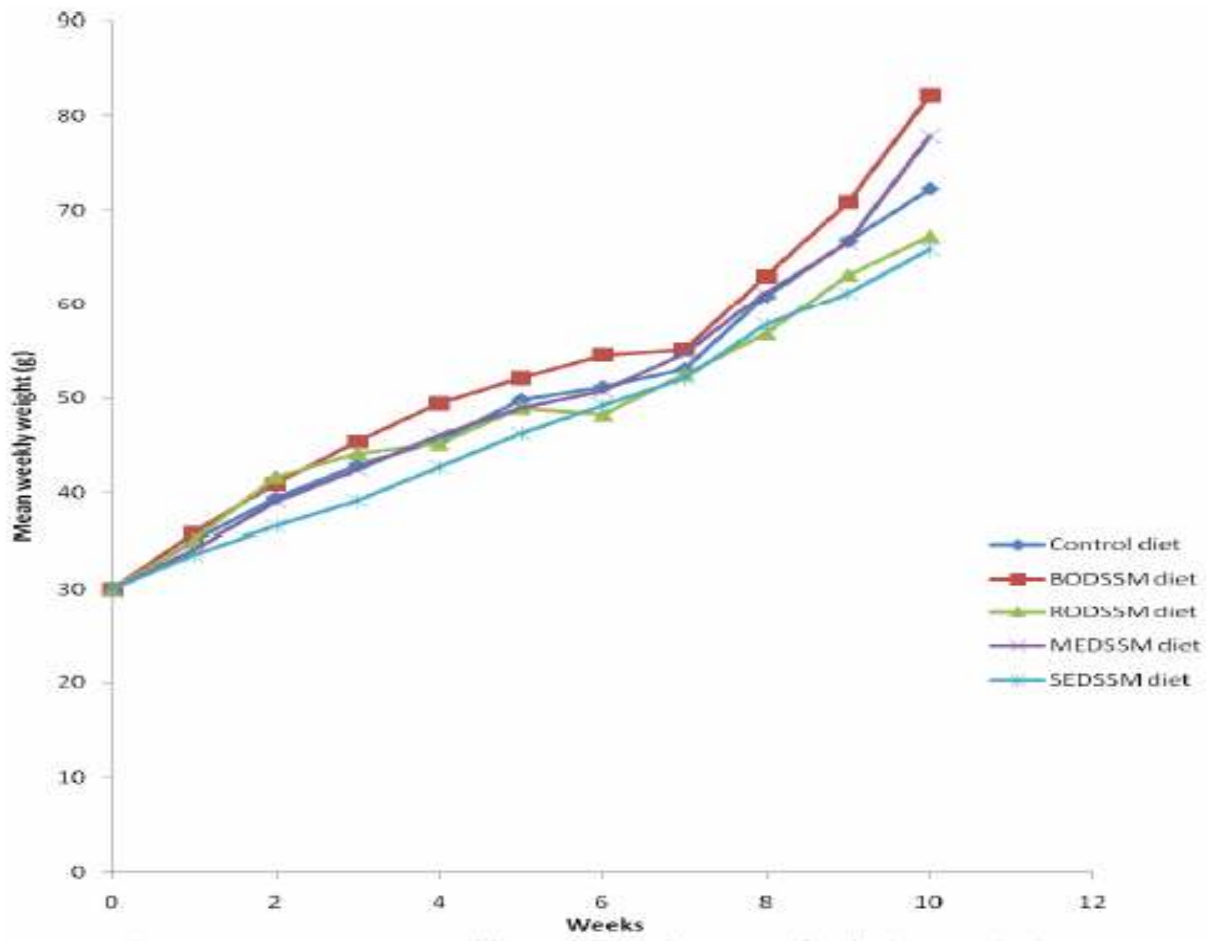
As observed in Table 5, mean weight gain was highest in fish fed diet boiled sunflower seed meal (52.16g) and lowest in those fed diet solvent-extracted sunflower seed meal (35.91g), Specific growth rate was highest in the fish fed diet boiled sunflower seed meal (0.62%) and lowest in those fed diet solvent-extracted sunflower seed meal (0.49%). FCR was highest in the fish fed diet roasted and solvent-extracted sunflower seed meal diets (0.62) and lowest in those fed diet boiled sunflower seed meal (0.47). These values were significantly different (p<0.05) among the dietary treatments and this is presented on table 5. Mean weekly weight changes of *Clarias gariepinus* juveniles fed diets including processed sunflower seed meal are presented in figure 1.

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	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

utilization (%)					
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

263 The letters *a,b,c,d,e*,etc indicate that mean values in each row with similar superscripts are
 264 not significantly different (*p*>0.05).
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 270 FIG. 1: Mean weekly weight changes of *Clarias gariepinus* juveniles fed diets including
 271 processed sunflower seed meal diets.
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3.5 Mineral compositions of *Clarias gariepinus* juveniles before and after the experiment

Mineral composition (sodium, calcium, potassium, magnesium, iron and phosphorus) were analyzed before and after the experiment are included in Table 6. The result recorded highest values of phosphorus were found in MSSM and the lowest in BSSM and RSSM, no significant difference ($p>0.05$) were observed among the treatments. Some minerals had lower values after the experiment when compared to the values recorded before the experiment (Table 6).

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

Parameter s	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 ^a _b	409.00±17.00 ^a	385.67±1.53 ^b	388.67±7.51 ^b	398.67±9.50 ^a _b	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 ($p>0.05$).

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3.6 Initial and final mean values of water quality parameters obtained during the 70 days feeding trials

The pH, dissolved oxygen (mg/Litre) and temperature ($^{\circ}\text{C}$) were measured throughout the experimental period are shown in Table 7. The three parameters the result shown that dissolved oxygen, temperature and pH ranged within very close limits. The final values significantly differed ($p < 0.05$) from the initial values. e and this was presented in table 7

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 ($^{\circ}\text{C}$)	27.05 $\pm$ 0.00	27.52 $\pm$ 0.02	27.53 $\pm$ 0.01	27.67 $\pm$ 0.02	28.01 $\pm$ 0.00	28.55 $\pm$ 0.01	27.72 $\pm$ 0.47
	Ph	6.81 $\pm$ 0.00	7.47 $\pm$ 0.05	7.46 $\pm$ 0.01	7.52 $\pm$ 0.02	7.51 $\pm$ 0.02	7.59 $\pm$ 0.01	7.39 $\pm$ 0.26
	DO (mg/l)	5.39 $\pm$ 0.03	5.42 $\pm$ 0.01	5.40 $\pm$ 0.00	5.39 $\pm$ 0.01	5.41 $\pm$ 0.00	5.40 $\pm$ 0.00	5.40 $\pm$ 0.01
BSSM	Temp ($^{\circ}\text{C}$)	27.07 $\pm$ 0.02	27.52 $\pm$ 0.03	27.52 $\pm$ 0.01	27.65 $\pm$ 0.02	28.12 $\pm$ 0.00	28.05 $\pm$ 0.05	27.66 $\pm$ 0.35
	pH	6.64 $\pm$ 0.03	7.53 $\pm$ 0.05	7.54 $\pm$ 0.04	7.56 $\pm$ 0.03	7.58 $\pm$ 0.02	7.56 $\pm$ 0.01	7.40 $\pm$ 0.34
	DO (mg/l)	5.41 $\pm$ 0.00	5.41 $\pm$ 0.01	5.40 $\pm$ 0.00	5.40 $\pm$ 0.00	5.39 $\pm$ 0.01	5.40 $\pm$ 0.00	5.40 $\pm$ 0.01
RSSM	Temp ($^{\circ}\text{C}$)	27.02 $\pm$ 0.01	27.52 $\pm$ 0.01	27.52 $\pm$ 0.03	27.65 $\pm$ 0.01	28.07 $\pm$ 0.00	28.05 $\pm$ 0.04	27.64 $\pm$ 0.36
	pH	6.83 $\pm$ 0.02	7.60 $\pm$ 0.00	7.61 $\pm$ 0.01	7.66 $\pm$ 0.01	7.64 $\pm$ 0.03	7.67 $\pm$ 0.02	7.50 $\pm$ 0.30
	DO (mg/l)	5.42 $\pm$ 0.01	5.40 $\pm$ 0.01	5.38 $\pm$ 0.02	5.38 $\pm$ 0.02	5.40 $\pm$ 0.00	5.41 $\pm$ 0.01	5.40 $\pm$ 0.01
MSSM	Temp ($^{\circ}\text{C}$)	27.06 $\pm$ 0.03	27.51 $\pm$ 0.04	27.50 $\pm$ 0.01	27.66 $\pm$ 0.02	28.06 $\pm$ 0.03	28.06 $\pm$ 0.00	27.64 $\pm$ 0.35
	pH	6.55 $\pm$ 0.05	7.83 $\pm$ 0.01	7.76 $\pm$ 0.03	7.77 $\pm$ 0.06	7.79 $\pm$ 0.01	7.84 $\pm$ 0.05	7.59 $\pm$ 0.47
	DO (mg/l)	5.41 $\pm$ 0.01	5.37 $\pm$ 0.02	5.38 $\pm$ 0.02	5.41 $\pm$ 0.01	5.40 $\pm$ 0.00	5.39 $\pm$ 0.02	5.39 $\pm$ 0.01
SSSM	Temp ($^{\circ}\text{C}$)	27.21 $\pm$ 0.00	27.51 $\pm$ 0.01	27.50 $\pm$ 0.05	27.67 $\pm$ 0.03	28.05 $\pm$ 0.01	28.05 $\pm$ 0.00	27.67 $\pm$ 0.31
	Ph	6.85 $\pm$ 0.02	7.80 $\pm$ 0.00	7.80 $\pm$ 0.01	7.79 $\pm$ 0.02	7.75 $\pm$ 0.03	7.76 $\pm$ 0.03	7.63 $\pm$ 0.37
	DO (mg/l)	5.41 $\pm$ 0.01	5.36 $\pm$ 0.03	5.39 $\pm$ 0.02	5.40 $\pm$ 0.01	5.40 $\pm$ 0.01	5.39 $\pm$ 0.02	5.39 $\pm$ 0.02

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 Udedibe and Nwaiwu (1987) 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 offal? and cotton seed,) by Gohl (1987), Oyelese (1994), Omitoyin (1995) and Alegbeleye (2005) 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 (Olaifa and Bello, 2011).

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 (Griffiths, 1991). Mukhopadhyay and Ray (1999) observed a reduction in the tannin content of sesame seed meal from 20g to 10g/kg after fermentation with lactic acid bacteria.

The values of oxalates obtained in the study support the report of Salunkhe *et al.*, (1991) who recorded reduces the oxalic acid content of the seed after dehulling. Oxalates reduce the physiological value of calcium in the seed (Narasinga, 1985; Johnson *et al.*, 1979. Also, the value of phytate obtained in this study was similar to the report of Smith (1968) who reported reduction in phytate of processed sunflower seed meal. The bio-

361 availability of phosphorus for animals seems to depend on the level of phytate-splitting
362 enzyme, phytase, in the intestinal tract.

363 Body protein levels increased in all fish, indicating that all diets supported fish
364 growth. Nevertheless, differences show that the whole body proximate composition was
365 influenced by the method of processing sunflower meals. This result was in support with the
366 report of Olaifa and Bello, (2011) who recorded increased in protein levels of *C. gariepinus*
367 juveniles fed diets differently processed African yam beans

368 There was general improvement in growth performance and nutrient utilization of
369 *Clarias gariepinus* fed diets differently processed sunflower seed meal. There were
370 significant variations ($p<0.05$) in the mean weight gain of fish fed the four diets differently
371 processed sunflower seed meal. These occurred as a result of the improvement of the
372 digestibility of the diets. This observation is similar to the findings of Farinu *et al.* (1992) and
373 Nwanna (2003) who also recorded significant mean weight gain in *C. gariepinus* with higher
374 digestibility values during their respective studies. Increase in weight gain in all experimental
375 treatments indicated that the experimental fish were able to convert the protein in the feed to
376 extra muscle in their bodies. The result of this study is in accord with the findings of Shabbir
377 *et al.* (2003) who reported higher growth of *Cirrhinus mrigala* fed diets including sunflower
378 meal. Similarly, Sahzadi *et al.* (2006) reported that hybrid fingerlings (*Catla catla* x *Labeo*
379 *rohita*) gained higher body weight and maximum total length when fed diets with sunflower
380 seed meal. The highest (0.62%) and the lowest (0.49%) values of specific growth rate
381 were recorded for boiled and mechanically-extracted sunflower seed meal diets respectively.
382 These values are slightly lower than the 0.904% and 0.668% recorded for *Cyprinus carpio*
383 fed fish meal-based and sunflower meal-based diets respectively (Khan *et al.*, 2003).
384 Different processing methods (BSSM AND MSSM) were observed to produce significant
385 differences ($p<0.05$) [LM2] in the values of specific growth rate when compare to the control
386 diets. Feed conversion ratio (FCR) was the highest (0.62) in fish fed RSSM and SSSM diets
387 and the lowest in those fed BSSM diet (0.47); this indicates a superior level of utilization of

388 the BSSM diet by the fish. The lower FCR indicates better feed utilization by the fish and is
389 justified by better growth performance of *C. gariepinus* fed BSSM diet. ANPU is a factor of
390 the digestibility, utilization and quality of the protein fed to fish (Adikwu, 2003). ANPU had
391 been linked to superiority of feed and its protein quality (Steffens, 1989). Therefore, fish fed
392 MSSM diet had the highest productive protein value (3.03) apart from the control (7.71).

393 Protein Efficiency Ratio (PER) is a measurement of protein effectiveness to provide
394 the essential amino acids needed by the fish as well as an index that had been associated
395 with fat deposition in fish muscles (DeSilva and Anderson, 1995). Protein efficiency ratio
396 (PER) showed significant differences among the treatments. Apart from the fish fed the
397 control diet which had a protein efficiency ratio (PER) of 1.06, the highest PER value (1.30)
398 was obtained in the fish fed BSSM diet and the least value (0.90) in those fed SSSM diets.
399 The best body weight gain, specific growth rate, feed conversion ratio, gross feed efficiency
400 ratio and nitrogen metabolism were recorded in *Clarias gariepinus* fed BSSM diet.

401 The survival rate of the control and SSSM treatments was 100% but survival in the
402 RSSM and BSSM treatments was only $96.67 \pm 1.67\%$. These values were closely related and
403 were not significantly different ($p > 0.05$) among the treatments. However, the robustness and
404 general well-being of the fish fed the sunflower seed meal diets are expressed by the
405 condition factor (K), which did not significantly differ from the control (Table 5). Also in terms
406 of biological parameters, the solvent-extracted sunflower seed meal diet produced the
407 lowest value, which could be due to the presence of thioglucosides. Anti-nutritional factors
408 such as tannin, oxalate acid, saponin, trypsin inhibitor, thioglucosides and phytate in
409 sunflower seed meals can reduce protein digestibility, thereby reducing the nutritive value in
410 relation to the availability of amino acids and growth performance (Aletor and Aladetimi,
411 1989).

412 Minerals are important for normal growth and serve as co-factors in several
413 metabolic reactions of animals including fish. A reduction of any of them from the diet will
414 negatively affect growth. In the present study, the processed sunflower seed meal diets

415 recorded significant ($p<0.05$) values of mineral deposition of sodium, calcium, potassium,
416 magnesium, iron and phosphorus. The highest value of phosphorus, iron, magnesium and
417 calcium were recorded in SSSM. Water quality parameters are given in Table 7 and were
418 within recommended limits for warm-water fishes (Boyd and Gross, 2000; Ajani, 2006).

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

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504 **COMPETING INTERESTS**

505
506 Authors have declared that no competing interests exist.
507

508 **AUTHORS' CONTRIBUTIONS**

509
510 Author A and B designed the study, author A supervised the study. Author B performed the
511 feeding trial and the statistical analysis as well as managed the literature searches. Author C
512 wrote the protocol, and wrote the first draft of the manuscript, managed the literature
513 searches. All authors read and approved the final manuscript.

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